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DETECTION OF THE LOOSE SMUT FUNGI IN EMBRYOS OF BARLEY AND WHEAT[1]

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The loose smuts of barley and wheat, caused by *Ustilago nuda* (Jens.) Rostr. and *U. Tritici* (Pers.) Rostr., respectively, although not usually of great economic importance have always been troublesome, especially to the seed grower. Both cause considerable loss when the incidence is high, and both are difficult to detect in seed samples and to control.

In Western Canada, a few fields of barley are reported each year which show infestations up to 20% of the plants. This represents, in most cases, a direct loss of about that amount. Usually, most fields show much less, but the annual surveys indicate that loose smut is nevertheless quite prevalent. The situation is much the same for wheat except that the infestations are usually less severe. Until recently, the barley loose smut records included both the true or internally-borne type (*U. nuda*), and the false or shallow-borne type (*U. nigra* Tapke). The discovery by Tapke (9) of a seedling-infesting loose smut of barley and his later work has cleared up many confusing questions in barley smut investigations.

The prevalence and sporadic occurrence of loose smut in both barley and wheat have been a source of considerable annoyance, rather than anxiety, to the seed inspector and seed merchant. Field inspections must be depended upon in efforts to determine true loose smut infections, and these are not wholly satisfactory.

TESTING SEED SAMPLES FOR LOOSE SMUT

To determine the percentage of loose smut in seed, the pathologist has usually resorted to a growing test. Greenhouse trials are fairly satisfactory but are time and space consuming. In greenhouse tests with smutty barley samples, a seed treatment with a mercury dust can be applied to eliminate the surface-borne smuts and so give a determination of true loose smut. Such a growing test has been conducted as a matter of routine on many barley samples by Dr. R. C. Russell of this Laboratory. In our first attempt (7) to outline a procedure for seed examination, a mature plant test was suggested for loose smuts as well as for barley stripe. Until recently the alternative to the growing test was to soak the seed,

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tease out the embryos, and fix, wash, dehydrate and embed them, following the usual histological technique. Sections were then made and stained. The method gives good results but is impracticable for routine examination.

There have not been many reports on routine methods for detecting loose smut infections. Contributions by Russian scientists, however, have thrown some light on the problem. Bubentzoff (2) developed a method for isolating *U. Tritici* from wheat seed; and, according to an abstract of the paper, it appears very promising. Saburova (6) states that loose smut infections of wheat can be determined by observing certain abnormalities in the ears of seedlings less than a month old. Later, Skvortzoff (8) used a maceration and staining technique on separated embryos to demonstrate infections by *U. Tritici.* More recently, Yablokova (10) reported on the application of fluorescent microscopy to the detection of *U. Tritici.* These four approaches deserve further study.

OBSERVATIONS ON SEPARATED EMBRYOS

From the start of cereal seed examination for disease as a routine procedure here, we have been confronted with the problem of detecting loose smut. During the past few years many attempts have been made to devise quick and reliable methods of diagnosis. The histological approach was explored and seemed promising, provided the embryos could be separated and handled in sufficient mass. Attention was directed therefore towards methods of embryo separation or removal. Maceration treatments were tried, and it was soon discovered that solutions of sodium or potassium hydroxide were surprisingly effective. It appears that these solutions cause a swelling of the endosperm starch coincident with a general softening and dissociation of the adhering hulls in barley and the pericarp and testa in both grains. This results in the release of the complete embryo without any noticeable distortion.

Embryo Separation

In most of our work, a 5% sodium hydroxide solution was used. About 30 grams of either barley or wheat seed is placed in a flask and covered with two or three times its volume of the solution. In earlier tests the seeds were pre-soaked in the ice-box overnight, but it is uncertain from later work whether or not pre-soaking is necessary. In 24 hours, free embryos may be seen in the soft gel-like mixture which has formed as the solution has acted on the kernels. These are easily detached by slight agitation upon the addition of water. To obtain more embryos, the mixture may be poured into a large dish and gentle pressure applied with a rubber stopper or some such instrument. A coarse screen will remove the larger fragments, leaving the embryos and smaller endosperm pieces. Finer screens may then be employed until a mixture of embryos and similar sized fragments is obtained. It may be necessary at this point to dip or pick the embryos out, and this was usually done. It is thought, however, that with suitable screening and other minor improvements the method would give a clean separation. Well over 50% of the seed usually release their embryos without much trouble, and commonly the yield is much better. It is believed that a yield approximating 100% can be expected with a little experience in the technique.

Detecting Infections

Two methods were employed in the examination of embryos to determine the extent of loose smut infection. In the first, or whole embryo method, the technique was kept very simple and with a minimum of manipulation the entire embryos were prepared for examination in the clearing fluid. In the second, or sectioned embryo method, the usual histological technique was employed, the embryos being embedded en masse in wax ready for sectioning. The whole embryo method seems particularly promising and is now described.

The Whole Embryo Method

As mentioned above, the embryos are separated and held in water. (a) They are allowed to remain in an excess of water for a time varying from several hours to overnight. The water may be changed once or twice to leach out most of the sodium hydroxide. (b) The water is drained off and 95% alcohol added. They may remain in this an hour or two. (c) The 95% alcohol is drained off and absolute alcohol added; this may be repeated to assure dehydration. (d) A portion of or the entire sample may then be placed in a syracuse watch-glass, the excess alcohol drained off, and cedar clearing oil added. Clove oil or other clearing solutions may be used but a rather thick cedar oil has been most satisfactory as it keeps the specimens in place when the dish is handled. Clearing begins immediately, and the embryos should be well cleared in an hour or so.

The scutellum is then examined carefully for the rather conspicuous infection foci. The somewhat dark brownish mycelium is easily seen in contrast with the transparent host cells (Figure 1). Frequently the entire scutellum is involved. Foci are commonly seen in the apical part of the scutellum, but almost as frequently they occur in lateral areas of the organ. The epithelium appears to be a favourite site, but the invasion also involves the adjacent parenchyma. The details of the infection can only be determined from sectioned and stained preparations; and these will be discussed later. For the gross examination as outlined in this method, a wide-field binocular was employed giving a magnification of about 70 times. Greater enlargement with a better instrument would no doubt facilitate the reading. Furthermore, it is an advantage if the specimens are arranged in groups or rows in the oil. A dish especially designed to allow the specimens to settle into rows or a dish marked into squares would make counting much easier. In cases of doubt, the embryo can be dissected and examined in greater detail.

Staining the whole embryo for examination needs further study, but the unstained preparations have proven surprisingly satisfactory.

The Sectioned Embryo Method

In this method, as mentioned above, the embryos are prepared for sectioning. Starting at step (b) as above, the specimens are transferred to 70 or 85% alcohol, then through the usual butyl-ethyl alcohol solutions to pure butyl alcohol. The specimens are left in the solutions for a period of about 2 hours. (c) Pieces of embedding paraffin are added to the vial containing the specimens in pure butyl alcohol and allowed to dissolve.

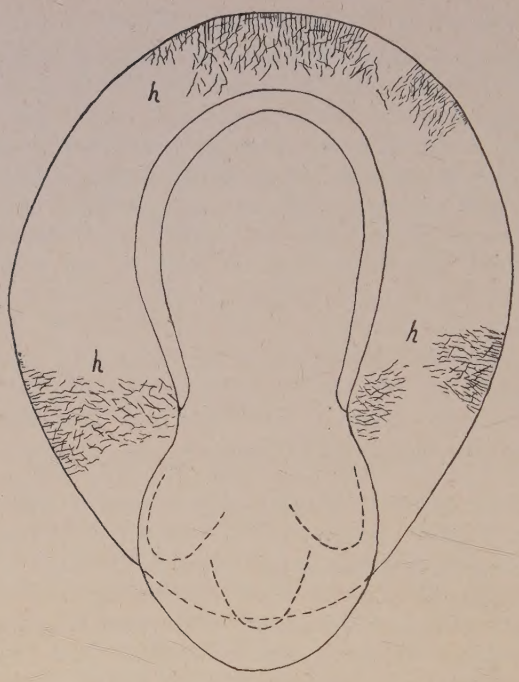

FIGURE 1. An outline sketch of a face view of a separated embryo of barley, showing the large scutellum in which foci of loose smut hyphae (h) may be seen after proper clearing. × 35.

This step is carried out in the wax oven held at 55° C. (d) As the paraffin dissolves, more is added until the specimens in a day or so are in pure paraffin. (e) The specimens are now ready to be transferred to the final embedding container. The size of the tray is quite important as the resulting block of wax should be large enough to contain as many specimens as possible in one plane and yet of a size convenient for cutting. Small porcelain trays (25 × 15 × 8 mm.) in which water-colours are sold were found satisfactory in this work. They were smeared very lightly on the inside with vaseline. To transfer the specimens, the excess wax is poured off and the mass of specimens dumped onto a clean piece of paper. In a moment they have cooled sufficiently to be picked up with a scalpel and placed in the tray, which is then put into the oven. If more wax is needed, it may be added. As the wax melts, the embryos settle to the bottom and upon gentle agitation arrange themselves in layers. In a tray of this size, each layer contains close to 100 embryos, the first layer of course being in the bottom of the tray. (f) The tray is next removed very carefully from the oven and cooled rapidly in ice-water. The block is easily removed and is ready to be placed on the microtome without much, if any, trimming. The block is secured with the bottom surface outward so the first layer of specimens is in position for sectioning. (g) A short ribbon can be taken off to assure contact with most specimens in the layer; then one cut or a short piece of ribbon is saved, and so on until enough material representing the first layer is obtained. The sections for staining are cut at 10μ. The instrument may be adjusted to 20μ or more to remove the remainder of the

first layer; then set at 10μ and specimens from the second layer taken. Likewise the third and additional layers may be sectioned. There is, of course, some overlapping between the layers. A block of the size mentioned will show for each stroke of the knife a group of sections representing as many embryos as are in the layer cut. Because of lack of orientation, sections will be of various sizes and angles, but most will show enough of the scutellum for the detection of the hyphae. Each paraffin section may be spread in the usual way separately on a slide or short ribbons may be used. After drying, the slides are ready for staining. (h) The wax is removed and the slides brought down to 50% alcohol, then placed in Harris' haemotoxylin for $\frac{1}{2}$ hour, washed with 50% alcohol, taken down to water, and placed in 5% aqueous solution of congo red for 3 hours. From here on the usual procedure is followed and the mounts are completed with balsam and cover-slip.

The slides are then ready for examination, and generally there is no difficulty in detecting the invaded embryos (Figure 2). The hyphae take the congo red fairly well, and many excellent preparations have been obtained. It is a simple matter to determine the percentage of embryos infected.

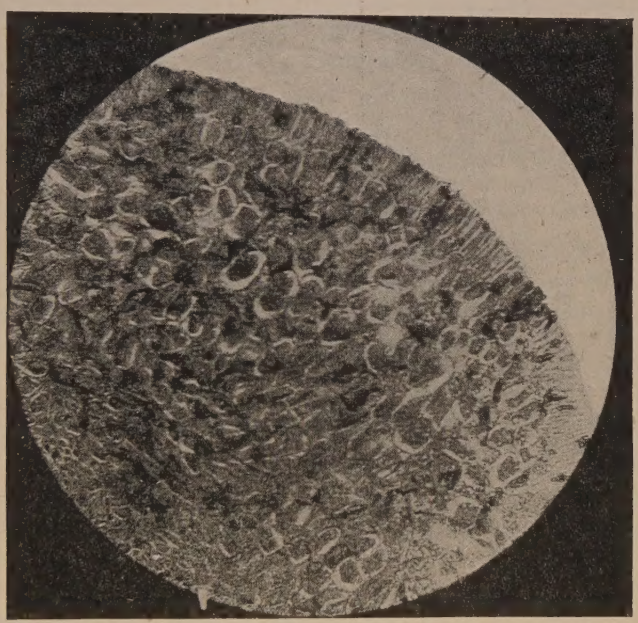

FIGURE 2. Showing a portion of the scutellum with the loose smut hyphae well established in the epithelium and parenchyma. From a section of a barley embryo. × 1000.

Comparative Data

It was difficult to get sufficient seed, especially of wheat, with known high loose smut content. Some samples were obtained, particularly of barley, upon which field notes had been made, and some were grown in the greenhouse for observation. The amount of *U. nuda* and *U. Tritici* was determined in several ways on the same samples (See Table 1).

TABLE 1.—THE AMOUNT OF TRUE LOOSE SMUT IN SAMPLES OF BARLEY AND WHEAT AS DETERMINED BY DIFFERENT METHODS

No.	Sample	Field notes	Greenhouse test	Embryos containing loose smut	
				Whole embryo	Sectioned embryo
		%	%	%	%
1	Glacier barley	21*	21	20	18
2	Reward wheat	4*	—	2	5
3	Reward wheat	—	—	1	—
4	Newal barley (Indian Head)	20*	14	14	11
5	Newal barley (Indian Head)	—	18	25	—
6	Newal barley (Edmonton)	—	10±	11	—
7	Newal barley (Swift Current)	—	—	8	—

* Counts based on smutted heads.

It will be seen that determinations by the various methods were in reasonably good agreement. The field notes were made earlier and for another purpose, and the percentage of smut was determined from head counts. In the greenhouse, around 200 plants were grown for smut counts and the percentage was based on the number of smutted plants. In the embryo examination, from 100 to 300 were examined for infection.

The sectioned embryo method, although somewhat more tedious than the whole embryo method, should be very exact and definite. The much faster and simpler whole embryo method, with some practice, should be just as reliable. A little experience would soon reveal varietal or other peculiarities, if any.

DISCUSSION

The main object of this study was to explore methods of examination for the determination of internally-borne loose smut infection in barley and wheat seed. The abundance of the hyphae in the scutellum was observed by Lang (3, 4) for both wheat and barley. He also mentioned a previous and similar observation by Hecke. Brioli and Schikkorra (1), by freehand and microtome sections, demonstrated the loose smut mycelium in the barley scutellum as well as just below the shoot bud. Lang, in describing the parts of the seed invaded, mentioned the heavy brownish mycelium in the embryo of the ripe seed. These characteristic hyphae and the foci of infections in the scutellum directed our attention particularly toward this organ. In the whole embryo method, these foci are quite easily demonstrated.

It was not part of this study to determine the mode of invasion and the final loci of the fungus in the seed. Nevertheless some observations in this connection were possible in the stained preparations of sectioned barley

embryos. There appeared to be no doubt of the heavy invasion of the scutellum, particularly in the area of the scutellar node from whence on the same level the fungus becomes established just below the growing point of the shoot. The scutellum sometimes appears completely invaded. In other specimens, there seem to be small areas involved in the lateral borders or at the tip or base of the scutellum. The epithelium is a favourite site of invasion, with the mycelium packed between the cells, but here and in the parenchyma there is considerable ramification apparently through some cells as well. There were no gross indications of injury; the staining reaction of host cells adjacent to hyphae appeared to be the same as distant cells.

Only one sample of Reward wheat seed was available for study and it did not contain a heavy infestation. The location of the fungus was in general the same as that in the barley embryos.

It was concluded that both methods have some promise as routine tests. There is still plenty of scope for minor modifications in the techniques, although it is thought that the fundamental principles are sound. Furthermore, it is believed that the examination of complete embryos might be applied to studies of viability, frost injury, and various other seed troubles. Preliminary attempts in staining *in toto* were not successful in this special study, but needless to say this phase deserves further study.

In barley seed it is quite important to know whether a surface-borne or an internally-borne smut or both are present in deciding what control treatment to recommend. The methods discussed should be useful in such problems.

SUMMARY

1. The problem of internally-borne loose smut of barley and wheat is reviewed briefly, especially in reference to methods used for the detection of these parasites in seed samples.

2. Two methods are outlined for detecting embryo infection. The embryos are removed by treatment with sodium or potassium hydroxide solutions. In the whole embryo method, they are dehydrated and cleared in cedar oil and examined without staining or sectioning; in the sectioned embryo method, the embryos are embedded, sectioned en masse, and stained. Both procedures gave reasonably good results.

ACKNOWLEDGMENT

The author wishes to acknowledge the helpful suggestions received from Mrs. Nebel and to thank her for examining some of the slides.

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THE PREVALENCE AND CONTROL OF SEED-BORNE DISEASES OF CEREALS IN MANITOBA[1]

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Some of the most destructive diseases of cereal crops (wheat, oats, barley, and rye) in Manitoba are caused by fungi and bacteria that are carried over from one crop season to the next either on the surface of the seed or within the seed. These diseases are commonly referred to as "seed-borne diseases." The seed-borne diseases of cereals of most common occurrence in this province can be grouped into three general classes: (1) smuts, (2) pre-emergence and seedling blights, and (3) leaf and stem spots. The chief crops affected by these are wheat, oats, and barley.

During the 10-year period ending 1937, the average area under field crops in Manitoba was about 6,219,000 acres. Of this acreage, the average sown to cereal crops was 5,616,000 acres (3). Cereals, therefore, occupied more than 90% of the land devoted to field crops. Wheat, of course, is the crop most extensively grown. Owing to the importance of cereal crops in this province, it is obvious that any group of diseases that seriously affects them is of great economic importance. Accurate estimates of losses in Manitoba due to seed-borne diseases of cereals are difficult to arrive at for the reason that information regarding the prevalence and severity of most of these diseases is too incomplete to provide an adequate basis for such estimates. It is generally recognized, however, that seed-borne diseases take a heavy annual toll from every important grain crop throughout the whole province. According to calculations made by Craigie (3), the average annual loss to Manitoba through the cereal smuts alone during the 22-year period 1916 to 1937 was $1,390,000. The seed-borne pathogens that are responsible for pre-emergence and seedling blights and for leaf and stem spots also cause substantial losses year after year in this province.

To obtain more definite information concerning the prevalence, severity, and distribution of seed-borne diseases of cereals in Manitoba, annual seed surveys were made from 1937 to 1942. Each year, from 400 to 1000 samples of cereal seed (wheat, oats, and barley) were examined critically for the presence of disease-producing parasites. The samples were obtained from seed stocks held on farms in Manitoba.

The principal objects of these studies were: (1) to develop reliable and practical methods for the pathological examination of cereal seed, (2) to ascertain the state of health of individual farmer's seed stocks, (3) to determine the fungus flora and the prevalence and severity of disease-producing fungi, (4) to study the relation of certain pathogenic fungi in wheat and barley seed to seed germination, and to the incidence of disease in the subsequent seedling stand, and (5) to determine the effect of seed treatment on the germination of healthy and diseased seed, and on the

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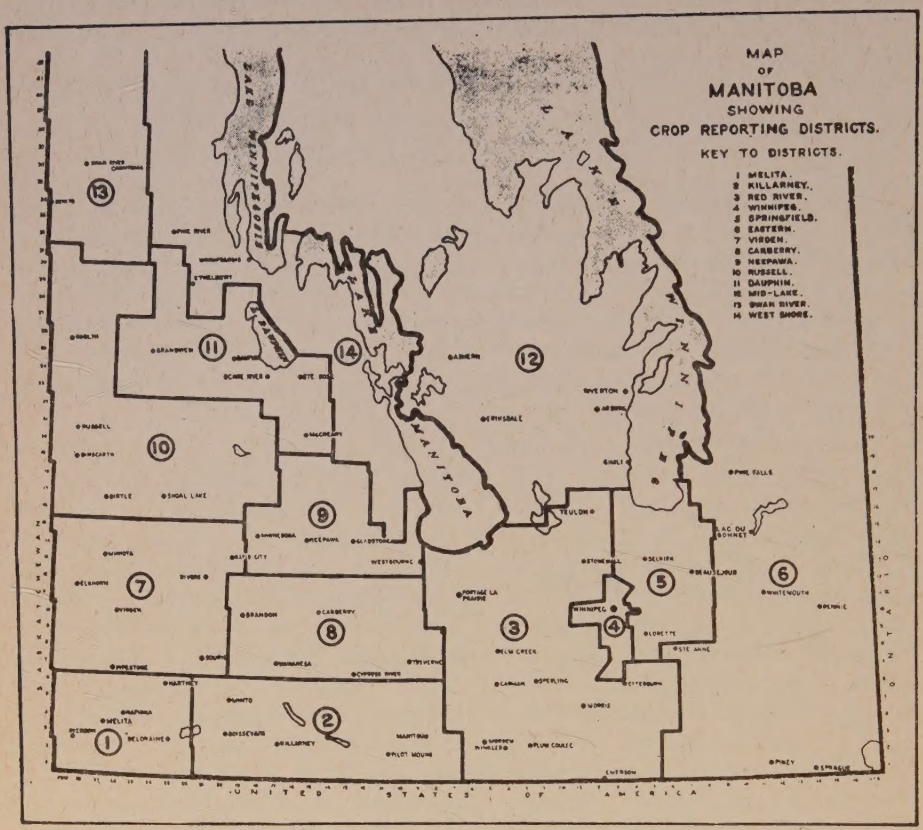

FIGURE 1. Crop reporting districts in Manitoba.

control of seedling-blight and leaf-spot fungi. A preliminary report on these studies has already appeared (6). A detailed account of them is presented in this paper.

MATERIALS AND METHODS OF PATHOLOGICAL SEED ANALYSIS

Samples of seed-grain from the 1937 to 1942 crops were obtained from individual farms in each of the major crop districts of the province (Figure 1). In 1941, for instance, about 40 farms, taken at random in each crop district, were visited. From these farms, 30 samples of wheat seed and 20 each of oat and barley seed were collected for disease examination. The total number of farms in Manitoba from which samples were obtained in 1941 was 520.

From a statistical standpoint, the taking of 520 samples may not constitute an entirely satisfactory sampling of the 56,000 farms in Manitoba, but for all practical purposes it was considered that the seed-grain samples examined in 1941, and in other years of the investigation as well, were representative of the seed stocks held on farms in this province.

In the first two or three years of the investigation, considerable time was devoted to a comparison of different pathological seed testing methods,

and to the development of new ones. The methods finally adopted to ascertain the state of health of each seed sample were: (1) a macroscopical examination of the sample to observe its physical condition and to detect the presence of ergot bodies, smudged kernels, smut balls, etc., (2) an agar-plate test to determine the internal fungus flora of the seed, (3) a microscopical examination of the seed washings to ascertain the spore load of smut and other fungi carried on the surface of the seed, and (4) a non-sterile soil test with disinfected and non-disinfected seed to measure seed germinability, to show the amount of seed-borne disease, and to indicate the response of each sample to seed treatment.

The agar-plate method used was as follows: 100 kernels, taken at random from each sample, were surface sterilized by immersing them in a solution of alcohol and mercuric chloride (1 part of 95% ethyl alcohol to 3 parts of 1 : 1000 mercuric chloride solution). The time of immersion was 4 minutes for wheat and rye, 2 for oats, and 5 minutes for barley. The kernels were then washed 3 times in sterile water. While they were being treated, acidified potato dextrose agar (pH 4.6) was poured into sterile Petri dishes and cooled to a temperature of about 50° C. By means of sterile forceps, the kernels were placed in the soft agar, 10 seeds per dish. The Petri dish cultures were then held at a temperature of from 24° to 26° C. for 8 days. At the end of this period, the organisms growing out from each kernel were either identified at once or isolated for further study and identification. The method described above is essentially similar to that developed and employed by the writers in earlier seed studies (5, 11). In the isolation of the pathogenic fungi found commonly associated with cereal seed, it has given satisfactory results.

The determinations of the smut spore load present on cereal seed samples were made by the method described recently by Cherewick (1). Briefly, it consisted of washing a portion of each sample, and of centrifuging and examining the wash water for the presence of smut and other fungus spores. By means of this test, it was found that a spore load of 1 : 128,000 (1 part of smut to 128,000 parts of seed, by weight) caused as high as 5% infection when the seed was grown under favourable conditions for infection.[3] In the present investigation, spore loads of this order were considered sufficiently high to necessitate seed treatment.

All samples of wheat, oat, and barley seed collected from 1937 to 1942 were grown in non-sterile soil in the greenhouse. Machacek and Wallace (13) reported that non-sterile soil is a satisfactory medium for testing seed germinability and certain seed-borne diseases of cereals. They found that the non-sterile soil test measures two important quantities of each seed lot, namely, (1) the percentage of seeds that are viable, and (2) the percentage of seeds that produce healthy plants. From the grower's standpoint, and in respect to a pathological analysis of the seed, the determination of the latter is the more important. When disinfected and non-disinfected seed is used, the non-sterile soil test also indicates the response of each seed lot to seed treatment.

[3] Unpublished data of Mr. W. Popp, Dominion Laboratory of Plant Pathology, Winnipeg, Man. The writers are indebted to Mr. W. Popp and Dr. W. J. Cherewick of this Laboratory for carrying out the smut tests reported in this paper.

Throughout the present investigation every effort was made to standardize and to employ procedures that would give a reliable estimation of the disease factor in the seed, and facilitate an accurate determination of other factors that are considered important in the appraisal of lots of cereal seed for seeding purposes. In other words, tests for seed-borne diseases were made in conjunction with tests of seed germinability. The more important results of the pathological seed tests described above are reported in this paper.

EXPERIMENTAL RESULTS

Seed-borne Diseases of Cereals in Manitoba

The seed-borne diseases of wheat, oats, barley, and rye of most common occurrence in Manitoba are listed in Table 1. Some of these, particularly the smuts, are of great economic importance, while others, although present almost every year, cause little damage in this province. In occasional years, however, some of the latter may cause appreciable loss.

Table 1.—List of the important seed-borne diseases of wheat, oats, barley, and rye in Manitoba, and their causal organisms

Crop, and common name of disease	Causal organism
Wheat	
Bunt, or covered smut	*Tilletia Tritici* (Bjerk.) Wint. and *T. laevis* Kuhn
Loose smut	*Ustilago Tritici* (Pers.) Rostr.
Seedling blight	*Helminthosporium sativum* P. K & B.
Spot blotch	*Helminthosporium sativum* P. K. & B.
Glume blotch	*Septoria nodorum* Berk.
Speckled leaf blotch	*Septoria Tritici* Desm.
Ergot	*Claviceps purpurea* (Fr.) Tul.
Bacterial black chaff	*Xanthomonas translucens* (J. J. & R.) Dowson amend. Hagb. f. sp. *undulosa* (S. J. & R.) Hagb. and f. sp. *cerealis* Hagb.
Basal glume blotch	*Pseudomonas atrofaciens* McCull.
Oats	
Covered smut	*Ustilago levis* (Kell. & Sw.) Magn.
Loose smut	*Ustilago Avenae* (Pers.) Jens.
Seedling blight	*Helminthosporium sativum* P. K. & B. and *Fusarium* spp.
Leaf blotch	*Helminthosporium Avenae* Eidam
Halo blight	*Pseudomonas coronafaciens* (Elliott) Stev.
Barley	
Covered smut	*Ustilago Hordei* (Pers.) Lagerh.
Loose smut	*Ustilago nuda* (Jens.) Kell. & Sw.
False loose smut	*Ustilago nigra* Tapke
Seedling blight	*Helminthosporium sativum* P. K. & B.
Spot blotch	*Helminthosporium sativum* P. K. & B.
Net blotch	*Helminthosporium teres* Sacc.
Septoria leaf blotch	*Septoria Passerinii* Sacc.
Ergot	*Claviceps purpurea* (Fr.) Tul.
Bacterial blight	*Xanthomonas translucens* f. sp. *hordei* Hagb. and f. sp. *hordei-avenae* Hagb.
Basal glume blotch	*Pseudomonas atrofaciens* McCull.
Rye	
Seedling blight	*Helminthosporium sativum* P. K. & B.
Ergot	*Claviceps purpurea* (Fr.) Tul.
Speckled leaf blotch	*Septoria Secalis* Prill. & Del.
Bacterial blight	*Xanthomonas translucens* f. sp. *secalis* (R. G. & J.) Hagb. and f. sp. *undulosa*

With the exception of ergot (*Claviceps purpurea*), the pathogens responsible for the diseases listed in Table 1 are carried either on the surface of the seed or within the seed. Some of these are distributed wholly in this way, while others may be transmitted in a number of additional ways. The fungus that causes ergot produces hard, horn-like bodies, called sclerotia, in the heads of diseased cereal plants. These bodies are carried with the seed but not attached to it. Ergot attacks rye chiefly, but also barley and wheat, and sometimes oats. Only an occasional sample of seed examined in the present investigation contained ergot bodies.

TABLE 2.—PERCENTAGE OF KERNELS INFECTED WITH *Helminthosporium* SPP., *Fusarium* SPP., AND OTHER FUNGI IN SAMPLES OF WHEAT, OAT, BARLEY, AND RYE SEED FROM THE CROPS OF 1937 TO 1942 IN MANITOBA

Kind of seed and year produced	Number of seed samples examined	Mean percentage of kernels infected with:*						Percentage of kernels fungus-free
		Helminthosporium spp.			*Fusarium* spp†	*Alternaria* spp.	Other fungi‡	
		sativum	*Avenae*	*teres*				
WHEAT								
1937	102	5.4	—	—	0.6	69.2	1.7	25.6
1938	290	4.4	—	—	0.4	74.3	1.7	20.8
1939	360	2.3	—	—	0.4	57.7	4.6	36.4
1940	390	2.9	—	—	1.0	71.7	2.4	23.8
1941	390	5.6	—	—	0.8	76.4	4.5	16.2
1942	178	4.8	—	—	0.9	76.7	2.8	16.9
1937–42	1710	3.5	—	—	0.6	70.2	3.2	23.9
OATS								
1940	127	1.2	0.6	—	1.1	51.6	3.1	44.2
1941	260	3.6	1.0	—	3.0	77.6	9.9	16.6
1942	131	2.6	0.7	—	2.0	72.1	3.7	23.9
1940–42	518	2.7	0.8	—	2.3	69.8	6.6	25.2
BARLEY								
1937	60	9.8	—	3.5	1.1	85.7	3.0	7.5
1938	60	7.6	—	1.3	1.7	89.0	4.3	6.4
1939	130	2.7	—	3.2	0.9	59.9	6.5	29.6
1940	219	3.4	—	3.1	1.4	81.7	3.3	12.1
1941	260	7.7	—	2.6	1.3	83.0	5.7	10.2
1942	161	5.2	—	2.2	1.8	80.8	2.5	13.3
1937–42	890	5.5	—	2.7	1.4	80.0	4.2	13.6
RYE								
1939–42	50	3.9	—	—	1.3	73.9	8.6	20.8

* Based on the results obtained by plating out 100 surface-sterilized kernels of each sample on potato dextrose agar in Petri dishes.

† Chiefly *Fusarium Poae*, *F. Scirpi* v. *acuminatum*, and *F. Equiseti*.

‡ Species of *Nigrospora*, *Curvularia*, *Cladosporium*, *Epicoccum*, *Septoria*, *Cephalothecium*, *Stemphylium*, *Pullularia*, *Penicillium*, and many other genera.

All the bacterial diseases of cereals known to occur in Manitoba are seed-borne[4]. Some of these, particularly halo blight of oats and black chaff of wheat, are common almost every year, and in certain seasons cause

[4] The writers are indebted to Dr. W. A. F. Hagborg, Dominion Laboratory of Plant Pathology, Winnipeg, Man., for information concerning the occurrence of bacterial diseases of cereals in Manitoba.

TABLE 3.—THE PREVALENCE IN MANITOBA OF *Helminthosporium sativum*, *H. Avenae*, *H. teres*, AND *Fusarium* SPP. IN SAMPLES OF WHEAT, OAT, AND BARLEY SEED FROM THE CROPS OF 1937 TO 1942

Kind of seed and year produced	Number of samples examined	*Helminthosporium sativum*		*Helminthosporium* spp.*		*Fusarium* spp.	
		Percentage of samples infected	Maximum percentage of kernels infected†	Percentage of samples infected	Maximum percentage of kernels infected†	Percentage of samples infected	Maximum percentage of kernels infected†
WHEAT							
1937	102	94	15	—	—	39	2
1938	290	87	29	—	—	27	3
1939	360	68	19	—	—	28	6
1940	390	85	24	—	—	56	25
1941	390	95	40	—	—	48	8
1942	178	91	50	—	—	54	7
1937–42	1710	84.2	50	—	—	41.0	25
OATS							
1940	127	62	5	41	9	50	20
1941	260	90	46	40	25	62	67
1942	131	77	26	31	15	60	24
1940–42	518	80.1	46	38.1	25	58.7	67
BARLEY							
1937	60	100	95	83	19	57	9
1938	60	93	44	43	15	68	9
1939	130	73	56	65	49	51	5
1940	219	87	27	59	29	46	20
1941	260	96	38	66	33	54	16
1942	161	87	33	57	22	71	12
1937–42	890	88.8	95	62.4	49	56.0	20

* *Helminthosporium Avenae* in oats, and *H. teres* in barley.
† Highest percentage of kernels infected in any sample.

appreciable loss. Unfortunately, no satisfactory method of detecting the presence of bacterial diseases in cereal seed has been developed. For this reason the studies reported in this paper deal mainly with the seed-borne diseases caused by fungi.

FUNGI ASSOCIATED WITH CEREAL SEED

The internal fungus flora of 3168 samples of cereal seed was determined by the plating method already described. The relative frequency of isolation of certain disease-producing, and other fungi, is given in Table 2. Fungi belonging to more than 60 genera were isolated. Those of most common occurrence belonged to the following genera. *Alternaria*, *Helminthosporium*, *Fusarium*, *Nigrospora*, *Curvularia*, *Epicoccum*, *Cephalothecium*, *Septoria*, *Pullularia*, *Cladosporium*, *Stemphylium*, *Penicillium*, *Aspergillus*, and *Mucor*. Several species of bacteria, yeasts, and actinomycetes were also isolated, but no distinctly pathogenic forms of these organisms were found.

As shown in Table 2, species of *Alternaria* were the predominant fungi. In the samples of wheat, oat, barley, and rye seed examined from 1937 to 1942, the average percentage of seeds infected with such species was, respectively, 70.2, 69.8, 80.0, and 73.9%. However, the writers (11) have shown that, although *Alternaria* spp. are commonly associated with Manitoba-grown cereal seed, and cause a discoloration of the kernel in wheat, barley, and rye, they are not responsible for seedling blight or leaf spot in the subsequent seedling stands. Thus, from a seed-borne disease standpoint, the presence of these fungi in such seed is not considered important.

Among the many other fungi isolated, the predominating pathogenic species were *Helminthosporium sativum* from wheat, barley, and rye; *H. Avenae* from oats; and *H. teres* from barley. Particular attention was paid to the isolation of these fungi, and the results of the soil infection tests, presented later in this paper, deal mostly with them. The prevalence of these fungi in samples examined from 1937 to 1942 is shown in Table 3.

It will be seen from Table 3 that *H. sativum* was present in 84.2% of the wheat, and in 88.8% of the barley samples. Although this fungus was found in 80.1% of the oat samples examined, it is not generally considered an important seed-borne pathogen of this crop. As might be expected, marked differences in the various crop samples occurred in respect to the amount of seed infection with *H. sativum*. In wheat, the infection range in the various samples was from 1 to 50%, and in barley from 1 to 95%.

In order to obtain a true picture of the importance of *H. sativum* in the samples, it is necessary to examine the frequency with which this fungus was isolated from individual kernels of the infected samples. It was found that an average of 3.5% of the wheat and 5.5% of the barley kernels cultured from 1937 to 1942 harboured this fungus. The average amount of kernel infection in rye and oat seed was, respectively, 3.9 and 2.7% (Table 2). It is evident that in Manitoba barley seed is always more heavily infected with *H. sativum* than is wheat seed, while oat seed carries the least infection.

H. sativum was widely distributed in Manitoba. The amount of infection in seed from the 12 major crop districts of the province is given in Table 4. As the data in this table show, wide differences occurred in the percentage of kernels infected with this fungus in different districts.

In the present study, the relation between the occurrence of seed infection by *H. sativum* and the normal rainfall during the growing season was given consideration. Rainfall data compiled by the Meteorological Division, Air Services of Canada, Winnipeg, Manitoba show that the normal rainfall during the period April 1 to September 9 at Brandon (Crop district 8), Portage la Prairie (Crop district 3N), Pierson (Crop district 1), and Russell (Crop district 10) was, respectively, 11.41, 11.12, 10.73, and 10.55 inches. The corresponding mean percentage figures for barley seeds infected with *H. sativum* were, respectively, 5.9, 7.4, 3.3, and 3.0% (Table 4). It is evident, therefore, that the heaviest seed infection occurred in districts with the highest rainfall during the growing season. This finding is in agreement with observations made in Manitoba over a long period of time and shows that severe infection of cereal seed with pathogenic fungi usually occurs in districts with the highest rainfall during the growing season.

TABLE 4.—THE FREQUENCY OF ISOLATION OF *Helminthosporium sativum*, *H. Avenae*, *H. teres*, AND *Fusarium* SPP. FROM WHEAT, OAT, AND BARLEY SEED PRODUCED IN DIFFERENT CROP DISTRICTS OF MANITOBA IN 1939, 1940, 1941, AND 1942

Crop district*	Mean percentage of kernels infected with:†							
	Helminthosporium sativum			*Fusarium* spp.			*Helminthosporium* spp.	
							H. Avenae	*H. teres*
	Wheat (95)‡	Oats (40)	Barley (60)	Wheat (95)	Oats (40)	Barley (60)	Oats (40)	Barley (60)
1. Melita	1.8	2.0	3.3	0.8	4.4	1.2	0.3	2.9
2. Killarney	2.8	2.3	4.5	0.8	3.1	1.8	0.3	2.0
3. Red River (S)	3.2	2.7	5.8	0.7	2.4	0.7	0.9	3.6
3. Red River (N)	4.6	3.6	7.4	0.7	1.3	1.3	0.8	3.8
4. Winnipeg	4.3	3.7	5.9	0.7	2.0	0.5	0.5	2.9
5. Springfield	4.7	3.2	4.6	0.6	0.6	0.6	0.7	3.4
6. Eastern	3.7	5.0	7.7	0.4	2.7	1.2	0.8	2.7
7. Virden	4.0	1.8	5.6	0.8	2.4	2.4	0.5	2.2
8. Carberry	4.5	2.2	5.9	1.0	1.8	0.9	0.7	2.9
9. Neepawa	5.0	2.3	4.5	1.0	2.2	1.3	0.6	2.7
10. Russell	2.9	1.8	3.0	1.4	1.9	3.0	1.0	2.2
11. Dauphin	4.5	3.0	4.2	0.5	0.6	1.0	1.8	2.5
13. Swan River	4.4	2.1	4.1	0.7	2.9	1.8	1.8	4.6

* Manitoba crop reporting district (see Figure 1). S = South, N = North.
† Based on the results obtained by plating out 100 surface-sterilized kernels of each sample on potato dextrose agar in Petri dishes.
‡ Approximate number of samples examined per crop district.

The average percentage of kernels infected with the leaf blotch fungus (*Helminthosporium Avenae*) in the 518 samples of oats examined was less than 1% (Table 2), while the maximum percentage in any sample was 25%. Nevertheless, this pathogen was harboured in 38.1% of the samples (Table 3). Internal infection of the kernels varied widely from district to district. The most severe infections of this type occurred in the cooler northern crop districts of Manitoba, namely, Dauphin and Swan River (Table 4). Additional plating tests with a large number of the samples of oat seed indicated that in certain samples infection of the kernels with *H. Avenae* was quite superficial, so that the organism was destroyed when the kernels were surface sterilized prior to being plated out. It must, therefore, be assumed that percentages of infection with *H. Avenae* higher than those recorded in Tables 2, 3, and 4 occurred in the oat samples.

An examination of 819 samples of seed barley collected from 1937 to 1942 showed that 62.4% of them carried the net blotch fungus (*Helminthosporium teres*). The maximum percentage of kernels infected in any sample was 49% (Table 3), whereas the average percentage in all samples was only 2.7% (Table 2). Generally speaking, barley seed produced in northern Manitoba (Swan River) was more heavily infected with *H. teres* than was that produced in other districts of the province (Table 4).

The frequency of isolation of species of *Fusarium* from the seed of wheat, oats, and barley from different crop districts is shown in Table 4. Obviously, these fungi are widely distributed on seed in Manitoba. Oat seed was more severely infected than was the seed of wheat or barley. In oats, the most severe seed infection occurred in the drier crop districts of the province, namely, Melita and Killarney.

In the present study, *Fusarium* spp., one or more, were isolated from 41.0% of the wheat samples, 58.7% of the oat samples, and 56.0% of the barley samples (Table 3). Although present in a fairly large proportion of the samples, *Fusarium* spp. were isolated from only a small percentage of the kernels. The average amount of infection in 1710 samples of wheat, 518 samples of oats, 890 samples of barley, and 50 samples of rye was, respectively, 0.6, 2.3, 1.4, and 1.3% (Table 2), while the highest percentage of kernels infected in any sample of wheat, oats, and barley, was, respectively, 25, 67, and 20% (Table 3).

Of a total of 16 species, varieties, or forms of *Fusarium* isolated, *Fusarium Poae*, *F. Equiseti*, and *F. Scirpi* var. *acuminatum* were the predominant species. The well-known pathogenic species *Fusarium culmorum* and *F. graminearum* were by no means commonly isolated. *F. culmorum* was isolated from only 16 out of 1710 samples of wheat, from 3 out of 518 samples of oats, and from 8 out of 890 samples of barley; while *F. graminearum* was isolated from 7 samples of wheat, 2 of oats, and 2 of barley. Most of the other species isolated are usually considered to be either non-pathogenic or only feebly pathogenic to cereals (8). Gordon (4) has given an account of the species of *Fusarium* commonly associated with cereal seed in Manitoba.

Beside the fungi already mentioned, certain species of *Septoria* pathogenic to cereal seedlings were isolated from the seed of wheat, oats, and barley. They were obtained most frequently from wheat, and least frequently from oat seed. Although the amount of injury from seed-borne *Septoria* spp. is relatively small in Manitoba, there is no guarantee that these species will always remain unimportant in this province. A report on the prevalence and importance of species of *Septoria* on cereal seed in Canada has been published by Machacek (10).

Relation of *Helminthosporium sativum* in Wheat and Barley Seed to Emergence and Seedling Blight

With a view to determining the relation between the percentage of seeds infected with *Helminthosporium sativum* and the amount of disease subsequently developing in the seedlings, the results of agar-plate and soil tests with 1481 samples of wheat and 747 barley samples from the crops of 1939, 1940, 1941, and 1942 were compared. The results of these comparisons are summarized in Table 5.

TABLE 5.—PERCENTAGE OF WHEAT AND BARLEY KERNELS INFECTED WITH *Helminthosporium sativum* IN RELATION TO THE PERCENTAGE OF KERNELS GERMINATING AND THE PERCENTAGE OF LESIONED SEEDLINGS. RESULTS OF AGAR-PLATE AND NON-STERILE SOIL TESTS WITH SEED SAMPLES FROM THE CROPS OF 1939, 1940, 1941, AND 1942 IN MANITOBA

Kind of seed and infection class	Percentage of kernels infected with *H. sativum**	Samples per class		Mean percentage plant emergence†	Mean percentage of seedlings with basal lesions†
		Number	Per cent		
WHEAT					
I	0 – 4	1047	70.7	92.2	2.4
II	5 – 9	303	20.5	89.9	6.9
III	10 – 14	87	5.9	88.0	10.3
IV	15 – 19	28	1.9	86.1	12.4
V	20 +	16	1.0	82.9	18.9
BARLEY					
I	0 – 4	455	60.9	93.4	1.4
II	5 – 9	172	23.0	94.4	2.2
III	10 – 14	81	10.9	89.2	3.9
IV	15 – 19	24	3.2	92.6	5.3
V	20 +	15	2.0	93.2	14.5

* As determined by agar-plate test with 100 surface-sterilized kernels of each sample.
† Results of infection tests in non-sterile soil with 100 kernels of each sample.

The data in Table 5 show that the percentage of wheat and barley seeds found by the agar-plate test to be infected with *H. sativum* was closely related to the percentage of seedlings with basal lesions when the seed was sown in non-sterile soil. An increase in the amount of seed infection was always accompanied by an increase in the percentage of blighted seedlings. Significant positive correlation coefficients of + 0.775 and + 0.841 were obtained for the data of the wheat and barley samples, respectively. These results confirm those of Christensen and Stakman (2), who found that the amount of seedling blight in barley was directly proportional to the percentage of seeds infected with *Helminthosporium* species, mostly *H. sativum*.

In barley, but not in wheat, there was a marked tendency for the percentage of seed infection to be higher than the percentage of seedlings with basal lesions. This difference was due, no doubt, to the fact that some lots of barley were infected with non-pathogenic or only weakly pathogenic strains of *H. sativum*. These results with barley emphasize the importance, in ascertaining the state of health of the seed of various cereal crops, of determining the relative prevalence of pathogenic and non-pathogenic strains of *H. sativum*, and of other fungi, in the seed.

As shown in Table 5, an increase in the percentage of wheat kernels infected with *H. sativum* was accompanied by a decrease in the percentage of seedling emergence. When the emergence and seed infection data were studied statistically, a significant negative correlation coefficient of −0.521 was obtained. These results indicate that seed-borne *H. sativum* is responsible for considerable pre-emergence blight in wheat. In barley, however, no significant difference in emergence was found between relatively

clean lots of seed (0 to 4% infection) and heavily infected samples (more than 14% infection). Thus, in barley, percentage emergence is not a reliable index of the degree of seed infection by *H. sativum*, and hence of the value of a sample for seeding purposes. The health condition of the seedlings arising from barley seed also must be taken into consideration.

With regard to the intensity of seed infection, the data of Table 5 show that in 91.2% of the wheat samples, and in 83.9% of those of barley tested, less than 10% of the seeds were infected by *H. sativum*. On the other hand, only in 1% and 2% of the wheat and barley samples, respectively, was the percentage of seed infection 20% or more. These results indicate that a large proportion of the wheat and barley seed being produced and sown in Manitoba is only lightly infected with this pathogen. From the grower's point of view this is important. The practical significance of relatively light infections with *H. sativum* in wheat and barley seed has yet to be determined.

Prevalence of Seedling-blight and Leaf-spot Fungi in Cereal Seed

To determine the amount of infection with the seedling-blight and leaf-spot fungi *Helminthosporium* spp., and probably *Fusarium* spp., in samples of wheat, oat, and barley seed, at least 100 seeds of each sample were planted in beds of non-sterile soil in the greenhouse. When the seedlings were 12 to 14 days old (second leaf stage) they were lifted from the soil, cleaned, counted, and examined for basal and leaf lesions. The percentage of seedlings with such lesions was used as an index of the health condition of each seed lot. The non-sterile soil test used in the present studies was that developed and employed by Machacek and Wallace (13).

On the basis of the soil test, the samples of wheat and oat seed were separated into 2 groups, one in which 5% or more of the seeds produced seedlings with basal lesions (diseased samples), and the other in which less than 5% of the seeds produced lesioned seedlings (healthy samples). Owing to the fact that the soil test alone is apparently unreliable for the appraisal of the amount of seed infection in barley with *Helminthosporium teres* (13), the barley samples were classified on the basis of the results of 2 tests—an agar-plate test and a soil test. In determining the health condition of a sample, the percentage of seed found by the agar-plate test to be infected by *H. teres* was added to the percentage of seed found to be lesioned (*H. sativum* and *Fusarium* spp.) when the seed was planted in non-sterile soil. If the sum of these figures exceeded an arbitrary figure of 5% the sample was classified as diseased.

As the results in Table 6 show, the average percentage plant emergence for the healthy samples of wheat, oat, and barley seed tested was 93.9, 95.8, and 94.5%, respectively; whereas the average percentage emergence for the diseased samples was 89.1, 85.6, and 92.7%, respectively. Thus, the presence of more than 5% seed infection with species of *Helminthosporium* and *Fusarium* in samples of wheat and oat seed reduced the percentage of seedlings that emerged from the soil by 4.8 and 10.2%, respectively. In barley, no significant difference in emergence was found between relatively clean lots of seed and lots that carried more than 5% seed infection with *Helminthosporium sativum* and *H. teres*.

TABLE 6.—THE RELATION OF SEED INFECTION WITH SEEDLING-BLIGHT AND LEAF-SPOT FUNGI (SPECIES OF *Helminthosporium* AND *Fusarium*) TO PLANT EMERGENCE IN SAMPLES OF WHEAT, OAT, AND BARLEY SEED FROM THE CROPS OF 1937 TO 1942 IN MANITOBA

Kind of seed and year produced	Number of samples examined	Diseased samples*		Healthy samples†	
		Number	Per cent plant emergence	Number	Per cent plant emergence
WHEAT					
1937	97	83	85.8	14	92.8
1938	88	44	91.9	44	94.4
1939	360	66	92.5	294	95.6
1940	390	66	91.1	324	91.5
1941	390	122	88.4	268	95.2
1942	151	35	85.4	116	93.3
1937–42	1476	416	89.1	1060	93.9
OATS					
1940	127	46	84.2	81	96.8
1941	260	23	86.6	237	95.5
1942	131	11	89.1	120	95.6
1940–42	518	80	85.6	438	95.8
BARLEY					
1937	60	56	88.4	4	90.0
1938	59	36	95.2	23	97.7
1939	130	11	96.7	119	95.8
1940	219	13	97.1	205	96.0
1941	260	25	92.9	235	92.6
1942	138	42	93.5	97	94.2
1937–42	866	183	92.7	683	94.5

* Samples in which 5% or more of the seeds, when planted in non-sterile soil, produced diseased seedlings.
† Samples in which less than 5% of the seeds produced diseased seedlings.

The prevalence of seedling disease caused by species of *Helminthosporium* and *Fusarium* in samples of wheat, oat, and barley seed examined in the course of the survey is shown in Table 7. According to the soil test 28% of the 1476 wheat samples, 15% of the 518 oat samples, and 40% of the 866 barley samples were diseased, while the average percentage of seedlings infected in these sets of samples was 9.4, 8.4, and 14.0%, respectively. These results indicate that in certain years a considerable amount of the seed of wheat, oats and barley that is produced in Manitoba is relatively free of seedling-blight and leaf-spot fungi, while in other years a high percentage of the seed may be severely infected with these disease-producing organisms. Observations made over a period of years have indicated that in Manitoba severe infection of cereal seed with these pathogens is usually associated with wet years, and more particularly with years in which wet weather prevails during the later part of the growing period.

In the course of the investigation the seed of several varieties of wheat, oats, and barley was examined for the presence of disease-producing fungi. It was found that some varieties were more susceptible to internal seed

TABLE 7.—PREVALENCE IN MANITOBA OF SEEDLING BLIGHT AND CERTAIN LEAF SPOTS CAUSED BY SPECIES OF *Helminthosporium* AND *Fusarium* IN SAMPLES OF WHEAT, OAT, AND BARLEY SEED FROM THE CROPS OF 1937 TO 1942

Kind of seed and year produced	Number of samples examined	Percentage of samples diseased*	Mean percentage of seedlings infected
WHEAT			
1937	97	83	15.1
1938	88	50	10.4
1939	360	18	7.3
1940	390	17	6.7
1941	390	32	8.2
1942	151	23	7.9
1937–42	1476	28	9.4
OATS			
1940	127	36	9.8
1941	260	9	6.5
1942	131	8	7.0
1940–42	518	15	8.4
BARLEY			
1937	60	98	20.4
1938	59	68	14.4
1939	130	32	9.7
1940	219	29	6.5
1941	260	33	8.3
1942	138	47	11.9
1937–42	866	40	14.0

* Samples in which 5% or more of the seeds, when planted in non-sterile soil, produced diseased seedlings. In barley the figures represent samples in which 5% or more of the seeds produced diseased seedlings and/or were infected with *Helminthosporium teres*.

infection by these fungi than were others. Thus, the variety grown may be an important factor affecting the prevalence of seed-borne diseases in Manitoba.

No attempt is made here to record the prevalence of retarded, stunted, deformed, or weak seedlings in the samples tested, or to discuss the probable causes of these troubles. The importance of such seedling abnormalities in the appraisal of cereal seed for seed purposes is fully realized, but a discussion of them lies outside the scope of this paper.

PREVALENCE OF SURFACE-BORNE SMUT IN SAMPLES OF CEREAL SEED

Samples of wheat and barley seed from the crops of 1939 to 1942, inclusive, and of oat samples from the crops of 1940, 1941, and 1942 were examined for the presence of smut. The present survey, however, deals only with those cereal smuts that are carried on the surface of the seed. They are as follows: bunt (covered smut of wheat), covered smut of barley, false loose smut of barley, covered smut of oats, and loose smut of oats. Although some progress has been made in developing a suitable technique for determining the presence of the loose smut fungi *Ustilago Tritici* and *U. nuda* in wheat and barley seed, respectively, no satisfactory practical

procedure is at present available. Here it may be mentioned that, although the loose smuts of wheat and barley are widely distributed in Manitoba, the annual loses to this province through them is considerably less than those caused by the surface-borne smuts (3).

TABLE 8.—PREVALENCE IN MANITOBA OF SURFACE-BORNE SMUT IN FARM SAMPLES OF WHEAT, OAT, AND BARLEY SEED FROM THE CROPS OF 1939, 1940, 1941, AND 1942

Kind of seed and year produced	Number of samples examined	Percentage of samples carrying:		
		Smut spores	Light smut-spore load*	Heavy smut-spore load†
WHEAT				
1939	360	46.1	44.2	1.9
1940	390	55.1	54.6	0.5
1941	390	78.9	73.5	5.4
1942	151	45.0	38.4	6.6
1939–42	1291	58.6	55.2	3.4
OATS				
1940	127	99.2	41.7	57.5
1941	260	100.0	14.6	85.4
1942	131	99.2	24.4	74.8
1940–42	518	99.6	23.7	75.9
BARLEY				
1939	130	100.0	46.2	53.8
1940	219	100.0	22.5	77.5
1941	260	100.0	21.9	78.1
1942	138	100.0	26.8	73.2
1939–42	747	100.0	27.3	72.7

* Spore load less than 1 : 128,000, by weight.
† Spore load at least 1 : 128,000, by weight. Seed treatment recommended.

The prevalence of the surface-borne smuts in the samples of wheat, oat, and barley seed examined in the course of the survey is shown in Table 8. Determinations of the smut spore loads present on the seed indicated that 58.6% of the wheat samples carried spores of bunt (*Tilletia laevis* and *T. Tritici*), 99.6% of the oat samples carried spores of covered smut and loose smut (*Ustilago Avenae* and *U. levis*), and 100% of the barley samples examined carried spores of the surface-borne smuts *Ustilago Hordei* and *U. nigra*. A point of greater importance is that the spore load was high enough on 3.4% of the wheat samples, 75.9% of the oat samples, and 72.7% of the barley samples, to make seed treatment for smut control seem necessary.

Spores of many different fungi were observed in the washings of samples of wheat, oat, and barley seed. In addition to those of smut fungi, spores of the following genera were of most frequent occurrence: *Alternaria*, *Helminthosporium*, *Fusarium*, *Epicoccum*, *Cladosporium*, *Puccinia*, *Penicillium*, *Aspergillus*, and *Mucor*. Bacteria, yeasts, and actinomycetes were also prevalent. For the most part the species of fungi observed were representative of the types commonly isolated from surface-sterilized cereal seeds (Table 2), and from soils in Manitoba (9).

Summary of Pathological Seed Tests

The results of the various pathological tests just presented for samples of wheat, oat, and barley seed from the crops of 1939, 1940, 1941, and 1942 in Manitoba are summarized in Table 9. On the basis of these tests the samples were separated into two classes, namely, diseased and healthy. In the diseased samples seed infection with surface-borne smut, or with other disease-producing fungi, was sufficiently heavy to necessitate seed treatment. As might be expected, and as the results in Table 9 show, many seed lots carried not only a smut load of that amount, but were infected to a dangerous degree with other disease-producing fungi as well. In the healthy samples, the seed was virtually free of smut and other disease-producing organisms. These samples were further sub-divided into those that germinated poorly (less than 91%) and those that germinated well (91% or above) in non-sterile soil. The germination of many of the former samples was not improved by seed treatment, and, consequently, they could not be considered entirely suitable for seeding purposes. In reporting on the health condition of the low-germinating samples to the growers concerned, an increase in rate of seeding was recommended.

Table 9.—Summary of tests made to determine the state of health of farm samples of wheat, oat, and barley seed from the crops of 1939, 1940, 1941, and 1942 in Manitoba

Kind of seed and year produced	Number of samples examined	Percentage of samples examined					
		Diseased*			Healthy†		
		Surface-borne smut	Seedling blight and leaf spots	Total	Seed of low germinability‡	Seed of high germinability*	Total
Wheat							
1939	360	1.9	18.3	18.3	18.9	62.8	81.7
1940	390	0.5	16.9	17.7	27.4	54.9	82.3
1941	390	5.4	31.8	35.4	21.3	43.3	64.6
1942	151	6.6	23.2	29.1	30.5	40.4	70.9
1939–42	1291	3.3	22.5	24.7	23.5	51.8	75.3
Oats							
1940	127	57.5	35.9	73.2	4.7	22.1	26.8
1941	260	85.4	8.8	88.1	6.5	5.4	11.9
1942	131	74.8	8.3	76.3	3.1	20.6	23.7
1940–42	518	75.9	15.4	81.5	5.2	13.3	18.5
Barley							
1939	130	53.8	31.5	66.2	1.5	32.3	33.8
1940	219	77.5	28.8	83.6	0.9	15.5	16.4
1941	260	78.1	33.3	81.9	6.6	11.5	18.1
1942	138	73.2	47.1	84.1	2.9	13.0	15.9
1939–42	747	72.7	34.3	80.1	3.3	16.6	19.9

* Samples in which infection with surface-borne smut, or with other disease-producing fungi, was sufficiently high to necessitate seed treatment. The seed of some of these samples was also of low germinability, and, consequently, increase in rate of seeding and seed treatment were recommended to the growers concerned.

† Samples virtually free of disease-producing fungi.

‡Percentage germination less than 91%. Germination not improved by seed treatment. Increase in rate of seeding recommended to the growers concerned.

§ Percentage germination 91% or more.

The data in Table 9 show that, on the basis of the arbitrary standards used, 24.7% of the seed samples of wheat, and more than 80% of the samples of oats and of barley examined required seed treatment for disease control. It is obvious, therefore, that a large proportion of the cereal seed used in Manitoba in 1940, 1941, 1942, and 1943 was not suitable for seed purposes. The results of the present survey support the view that all farmers in Manitoba should subject their cereal seed, particularly their oat and barley seed, to surface disinfection, unless it has been found by proper examination to be free from disease-producing organisms.

Control of Seedling Blight and Certain Leaf Spots of Cereals by Seed Treatment

In the course of the present seed-borne disease investigation, seed treatment tests were made with a large number of both diseased and healthy samples of wheat, oat, and barley seed. For these tests, two 100-kernel lots were taken at random from each seed sample. One of these was treated with a dust disinfectant consisting of one part of Ceresan (5% ethyl mercury phosphate) to two parts of talc. To insure complete coverage of each kernel the dust was applied in excess. The second 100-kernel lot remained untreated. The treated and untreated lots were planted in adjacent rows in beds of non-sterile soil in the greenhouse, and notes were taken when the seedlings were in the 2-leaf stage. The results are given in Table 10.

The data for the diseased seed samples in Table 10 show that treatment of infected seed of wheat, oats, and barley with an organic mercury dust was consistently beneficial to emergence, and gave almost complete control of seedling blight and leaf spots caused by species of *Helminthosporium* and *Fusarium*. The higher the degree of seed infection the more beneficial was the treatment. Furthermore, it was found that lightly infected cereal seed, when properly treated with a mercury dust, was just as good for seeding purposes as was healthy seed. Of course, healthy seed is preferable if it is available, but unless the seed is known to be healthy treatment is recommended.

The mean percentage germination figures for the healthy seed samples of wheat, oats, and barley examined (Table 10), indicate that the germination of such samples was not increased significantly by seed treatment. This finding supports the view that there is little value in treating healthy seed of cereals, unless it is sown in heavily infested soil.

That it is a mistake to neglect treating even lightly infected seed of small grain crops is shown by the present greenhouse studies. The fact that a large proportion of the cereal seed produced and sown in Manitoba carries disease-producing organisms indicates that seed treatment should be more widely and consistently practised in this province. The results of the present greenhouse tests are in agreement with those found in extensive field trials in Manitoba (7, 11, 12), and show that some of the most destructive seed-borne diseases of cereals can be effectively controlled by seed treatment.

TABLE 10.—THE EFFECT OF TREATING DISEASED AND HEALTHY SAMPLES OF WHEAT, OAT, AND BARLEY SEED WITH CERESAN ON PLANT EMERGENCE, AND ON THE CONTROL OF SEEDLING BLIGHT AND CERTAIN LEAF SPOTS. (DATA ARE MEANS OF SAMPLES EXAMINED FROM THE CROPS OF 1939, 1940, 1941, AND 1942)

Kind and condition of seed	Number of samples examined	Percentage plant emergence			Percentage of seedlings diseased		
		Treated seed‡	Untreated seed§	Increase due to treatment	Treated seed‡	Untreated seed§	Increase due to treatment
WHEAT							
Diseased*	622	93.4	87.3	6.1	0.3	5.1	4.8
Healthy†	669	95.9	94.6	1.3	0.1	0.1	0.0
OATS							
Diseased	207	88.7	84.5	4.2	0.3	4.7	4.4
Healthy	311	96.0	95.9	0.1	0.2	0.3	0.1
BARLEY							
Diseased	213	91.3	88.1	3.2	0.1	5.0	4.9
Healthy	534	96.8	96.9	−0.1	0.1	0.3	0.2

* Seed samples in which less than 91% of the untreated seeds produced healthy seedlings.
† Seed samples in which 91% or more of the untreated seeds produced healthy seedlings.
‡ Seed dusted with dilute Ceresan (5% ethyl mercury phosphate).
§ Natural, untreated seed.

DISCUSSION

The standards used to determine the value of seed-grain for seeding purposes are based on purity of variety, freedom from weed seeds, and germination. Thus, high value is attached to the ability of the seed to germinate and produce strong plants. One of the most important factors influencing the germinability of cereal seed is the presence of disease-producing organisms in or on the seed. The present studies have indicated clearly that the presence of more than 5% kernel infection with these organisms markedly reduces the germination of wheat and oat seed. Furthermore, the planting of infected seed practically ensures the perpetuation of the disease. Infected seed carries the disease into areas where it did not previously exist. Unfortunately, much of the seed of wheat and oats and some of the barley seed that is produced and sown in Manitoba carries with it organisms that reduce germinability. Such seed is of inferior quality, and should be subjected to surface disinfection before it is used for seeding purposes.

The most important seed-borne diseases of small grain crops in Manitoba are the surface-borne smuts of oats and barley. Because of the marked prevalence of these diseases in this province, it is important that all practical means be employed to suppress them. Perhaps one of the most effective ways of doing this is to examine the seed for the presence of smut spores before it is used or sold for seeding purposes. Such an examination would indicate whether or not seed treatment could be employed with profit. Furthermore, if the seed was found to be free from smut its value for seeding purposes would be enhanced.

The grower, usually, is unable to distinguish between seed that carries disease and seed that does not. Testing seed for the presence of disease has to be done by qualified technicians in properly equipped laboratories. Unfortunately, the facilities needed to examine individual farmers' seed samples for smut, and other diseases, are not available. At the present time, the testing of cereal seed for the presence of disease parasites is confined to high-grade seed, that is, to stocks of Foundation, Elite, and Registered seed. The results presented in this report indicate that the establishment of a more adequate pathological seed service would constitute an important step forward in the interests of seed-borne disease control and of good seed production in Manitoba.

In view of the fact that 25% of the seed stocks of wheat and more than 80% of those of oats and of barley produced and sown in Manitoba have been found diseased, all farmers in this province would be well advised to treat their cereal seed each year as a routine practice. In other words, unless the seed is known to be healthy, treatment is strongly recommended. In addition to destroying seed-borne pathogens, seed treatment with an approved disinfectant may offer considerable protection against pathogenic organisms already present in the soil. Protection against these is only likely to be effective until shortly after the seed has germinated, but this is the critical period when such soil-inhabiting organisms are most likely to reduce germination and plant emergence. Seed treatment, in spite of the need for safer and better fungicides and for more efficient seed-treating machinery, seems to offer more practical and tangible results than any other procedure available for preventing the losses caused by seed-borne diseases of cereals in this province.

The examination of numerous samples of cereal seed from all the major crop districts of Manitoba has shown that each year a very large proportion of the samples carried disease-producing organisms (smut fungi, seedling-blight fungi, leaf-spot fungi, etc.). Not all Manitoba grown seed, however, carries seed-borne disease. As a rule, cereal seed produced in dry districts is less likely to be diseased than seed produced in wetter districts, but the variability of moisture is so great from season to season in any district that a distinction between dry and wet districts is too ill-defined to be applied in respect to the prevalence of seed-borne diseases.

Although the present seed studies have dealt almost entirely with seed-borne diseases of cereals caused by fungi, the importance of those of bacterial origin is fully recognized. However, further seed investigations are needed before the prevalence and importance of bacterial pathogens in cereal seed can be accurately determined. Another aspect of the cereal seed disease problem that has received far too little attention in this survey is that dealing with mechanical, frost, and other kinds of seed injury. The importance of these seed troubles in the production of grain for seed purposes is fully realized.

SUMMARY

With a view to ascertaining the state of health of the seed of wheat, oats, and barley being grown in Manitoba, more than 3000 farm samples of seed from the 1937 to 1942 crops from all over the province were examined.

Plating tests of the samples showed that several kinds of organisms, particularly fungi, were associated with much of the seed. Of the fungi isolated, *Helminthosporium sativum* on wheat, barley, and rye, *H. Avenae* on oats, and *H. teres* on barley were the predominating pathogens. Several species of *Fusarium* were frequently isolated, particularly from oat seed, most of which were either non-pathogenic or only feebly pathogenic to cereals. The well-known pathogenic species *Fusarium culmorum* and *F. graminearum* were by no means commonly isolated.

Extensive infection tests in non-sterile soil have indicated clearly that a close positive relationship exists between the percentage of wheat and of barley seeds infected by *H. sativum* and the occurrence of disease in the subsequent seedling stands. In wheat, but not in barley, seed infection with this fungus was associated with low germination.

Samples of cereal seed from the crops of 1939, 1940, 1941, and 1942 were examined for the presence of smut. Of the 1710 wheat samples examined, only 3.4% carried more than a trace of bunt (*Tilletia Tritici* and *T. laevis*). A total of 75.9% of the 518 samples of oats examined carried spores of loose and covered smut (*Ustilago Avenae* and *U. levis*) in sufficient amounts to make seed treatment necessary. The spore load of covered smut and false loose smut (*Ustilago Hordei* and *U. nigra*) on 72.7% of the 747 seed samples of barley examined was sufficiently heavy to necessitate seed treatment. These results indicate that seed treatment, particularly of oat and barley seed, is not being adequately carried out in Manitoba.

The amount of infection caused by smut fungi, seedling-blight fungi, and leaf-spot fungi on seed grain was found to vary appreciably from district to district, and from year to year, depending largely upon the particular environmental conditions under which the seed was produced. The survey indicated that some districts in Manitoba are more desirable for the production of healthy seed of cereals than are others.

Field observations made over a period of years in Manitoba have shown that, if climatic conditions favour early ripening and harvesting of cereal crops, the incidence of infection by seed pathogens is low; but, if warm humid weather occurs during ripening and harvesting, the incidence of infection is high. Another factor affecting the prevalence of seed infection with disease-producing organisms is the variety grown.

In greenhouse tests, treatment of a large number of infected samples of wheat, oat, and barley seed with an organic mercury dust gave almost complete control of seed-borne disease caused by species of *Helminthosporium* and *Fusarium*. Seed treatment improved the germination of infected wheat and oat seed, but it had little or no effect on the germination of healthy cereal seed. The treatment, therefore, of healthy seed is of little value unless the seed is sown in heavily infested soil.

The present survey shows that the health condition of the seed of wheat, oats, and barley being produced and used annually in Manitoba is by no means satisfactory. Almost 25% of the seed stocks of wheat and more than 80% of those of oats and of barley examined from the crops of 1939, 1940, 1941, and 1942 carried disease-producing organisms in sufficient amounts to necessitate seed treatment.

The most important seed-borne diseases of cereals in Manitoba are the surface-borne smuts of oats and barley. In the absence of adequate facilities for determining whether or not individual farmers' seed stocks of oats and barley are free from smut, and other diseases, it is strongly recommended that all seed of these crops produced in Manitoba should be treated with an approved disinfectant before it is sown.

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CLIMATIC FACTORS AFFECTING CROP PRODUCTION

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The settlement of Northern areas is largely dependent on the climatic conditions encountered. The weather not only determines the choice of crops to be grown but it also dictates how these crops are to be handled.

The Dominion Experimental Station at Beaverlodge has on record perhaps the largest body of meteorological data of any of the Northern Stations, and is situated on what might be regarded as the Southern fringe of Northern agriculture. For this reason the Beaverlodge data will be used to a considerable extent in this survey.

Precipitation

Precipitation records are available at Beaverlodge for 29 years. The average annual precipitation is 17.43 inches, 6 inches coming in the form of snow. The average monthly winter distribution is fairly regular, while spring opens with 0.80 inch of moisture in April. May precipitation amounts to 1.56 inches, June 2.11 inches and July 2.29 inches. Commencing with August the amount drops from 1.85 inches in that month to 1.15 inches in October. It is significant that in the 29-year period the April to August precipitation falls below 7 inches in 8 years and is in excess of 10 inches in 6 years.

By comparison Lacombe reports slightly less annual precipitation than Beaverlodge, with 3.3 inches coming in the winter months and 11.6 inches in the open season. The extra 3 inches of summer precipitation comes at a very opportune time.

At Fort Vermilion the annual precipitation is 11.90 inches, 7 inches of it coming in the summer months. At Norman, on the Mackenzie, the summer precipitation is about 6 inches, and beyond the Arctic circle it runs from 4.5 to 5 inches.

Evaporation

Records of evaporation from a free-water surface are available at Beaverlodge for 23 years. For the most part the tank was in operation from about April 20 to the end of October. The average yearly evaporation is 18.44 inches but in individual seasons it ranges from 11 to 22 inches. For the period during which the tank was in operation the precipitation averaged 11.12 inches or 7.32 inches less moisture than was evaporated from a free-water surface. The evaporation-precipitation ratio was lowest in the wet summer of 1935 and highest in the dry summer of 1922. By months, there was rather more evaporation in July than in May, June, or August, with a definite decrease in September and a further decrease in October.

While the May to September evaporation at Beaverlodge, in a 21-year average is 16.93 inches it is reported as being 15.39 inches at Lacombe, 24.60 inches at Lethbridge, and 33.17 inches at Manyberries for about the same period.

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Air Temperature

There is very little difference in the reported mean annual temperature at Beaverlodge and Lacombe. The Beaverlodge mean of 35.6 degrees is actually higher than that recorded at points in Manitoba and Saskatchewan with the exception of Morden and Swift Current. This condition results, however, from the December to March average at Beaverlodge being 13.9 degrees, while six Manitoba and Saskatchewan points average 6.4 degrees. The Beaverlodge average for April is slightly in excess of that of Melfort and Scott. The May mean stands at 47.9 degrees at Fort Vermilion, 49.1 degrees at Beaverlodge and 52.2 and 53.3 degrees at Swift Current and Manyberries, respectively. In June, Beaverlodge averages 55.5 degrees, Fort Vermilion 55.9 degrees, Lacombe 56.1 degrees and Manyberries 60.4 degrees. July temperatures average about 5 degrees higher throughout, followed by a decrease in August. In September the readings are about the same at all points as in May. The Beaverlodge readings are taken on a hillside which accounts for some modification as compared with ruling temperatures of the region.

Soil Temperature

The soil temperature at 3 inches drops to 24.36 degrees in January. In February it climbs to 25.12 degrees, in March to 27.73 degrees, in April to 35.42 degrees and in May to 48.31 degrees. In June it averages 56.79 degrees, in July 62.11 degrees and in August 59.25 degrees. Thus it is well into May before soil temperatures are sufficiently high to permit nitrification.

Sunshine

Beaverlodge enjoys 2103 hours of bright sunshine per year. Fort Vermilion has slightly less, 2093 hours, while Lacombe has 2170 and Lethbridge 2348 hours. A trend such as this seems reasonable, but its harmony is challenged by the Swift Current total of 2095 hours. Beaverlodge sunshine, as registered, is comparatively low until April, when it averages about 6 hours more than does Lacombe. In May the spread is increased to 26 hours, while in June it is 8 hours less than at Lacombe. In July the record is even, after which Lacombe is in the lead. At Fort Vermilion, however, the August sunshine is still in excess of that of Lacombe. It is known, however, that many hours of dull light are not recorded, which limits the value of these readings. In final analysis we must observe the reaction of plant growth to the sunlight to get the full meaning of this factor. Some data are available but many more will be needed before the North can be cropped to full advantage.

Wind

The average wind velocity at Beaverlodge is 8.3 miles per hour. This ranges from about 7 miles in the winter months to a peak of 10.5 miles in May. This is unfortunate as spring-sown crops are then vulnerable and provide the least cover. The highest wind velocity recorded at Beaverlodge is 48 miles per hour. Some rather serious soil drifting has occurred where preventive measures have not been taken.

DISCUSSION

On the Beaverlodge Station Marquis wheat, on fallow, has averaged 36.5 bushels per acre over a 30-year period. On the other hand Grimm alfalfa makes an average of 1½ tons per acre from the one cutting usually taken. There is usually ample surface moisture for seeding, followed by a period rather unfavourable for growth. Plants which have become established develop a strong root system in this period, which serves in good stead later. Late-sown crops or crops sown on land where moisture-saving practices have not been followed may not do so well. June is sometimes dry and too frequently September and October weather makes threshing catchy.

Wheat seems to stand dry spells better than oats, while barley and flax are much inferior in this respect. These latter crops have been tried time and again but have not become popular. The leading forage crops are brome, alfalfa and sweet clover. These are readily established in strong stands but their production in hay or pasture is not equal to that of wheat or oats. On the whole there is not sufficient early-summer moisture for the growing of timothy, red clover, or alsike. Winter wheat does better at Beaverlodge than at Edmonton, but not as well as in Southern Alberta. The 17-year average yield of Kharkov 22 is 28.7 bushels per acre. In some years killing is attributed to chinook influence, which bares knolls and causes icing in low spots.

It is common practice to fallow one year in four. Rotations should include the use of a forage crop about three years in eight if fibre is to be maintained. Annual and winter annual weeds are particularly troublesome and for the most part the soil tends to blow and wash readily.

The low soil temperature in the early summer, together with the comparatively low precipitation at that time, is not conducive to the rapid formation of soil nitrates. Sod should be broken early if a satisfactory grain crop is expected on the land the following year and no case has been made for cover crops on summerfallow. Winter wheat will turn yellow in late May unless there are surface rains to promote nitrification. This condition may possibly apply in greater degree further North since on the whole the soil in those areas is not particularly rich and the rainfall is low.

Hopkins and Leahey report the average date of the first seeding of wheat as April 28 at Beaverlodge and Lacombe, April 21 at Lethbridge and May 1 at Fort Vermilion. Corresponding harvesting dates are August 25, 26, 8 and 21. In this regard attention is drawn to the report of Albright and Stoker which indicates local incidence in the frost factor.

And, finally, a word about the climate in general. In its natural state there is a fairly complete cover of poplar, willow and spruce, along with other associated species and intermixed with this is a tall growth of grasses and legumes. As breaking takes place the settlers find plenty of moisture for their crops, a generous supply of nitrogen, and a very definite frost hazard because of limited air drainage. As areas open up these factors become moderated and to-day combine harvesting is being considered seriously in areas where crops used to be cut in early September, whether or not they were mature. This earlier harvesting indicates a drier soil climate and emphasizes the need for moisture conservation.

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ASCORBIC ACID CONTENT OF TOMATO VARIETIES AND ITS RETENTION IN PROCESSED PRODUCTS[1]

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Tomatoes are one of the commonly available but variable sources of ascorbic acid. Canned tomatoes and tomato juice are the most important natural sources of ascorbic acid being manufactured in volume in Canada at a reasonable cost to consumers. If properly grown and prepared, these products should be not only attractive and palatable but also a good source of ascorbic acid. Reports published in April, 1944, and in December, 1944, by the Combined Food Board on food consumption levels in the United States, Canada and the United Kingdom showed that in Canada the only vitamin supply seriously deficient was ascorbic acid.

The outstanding value of the tomato in nutrition is due in great measure to its ascorbic acid (vitamin C) content. Published analyses indicate the extreme range that may be found in ascorbic acid content of tomatoes from various sources and at different times of the year. This range is from 5 to 50 milligrams per 100 grams of fresh material. However, the normal range of commercially grown summer varieties is probably 15 to 30 milligrams.

REVIEW OF LITERATURE

There are several published accounts on the ascorbic acid content of varieties and strains of tomatoes, particularly as they occur in the United States. Also a number of investigators have reported on several factors which may be responsible for fluctuations in ascorbic acid content. Excellent reviews on the subject are given by Hamner and Maynard (9), and Maynard and Beeson (17), and corroborated by more recent works. It is apparent that the principal factors affecting ascorbic acid value of tomatoes are variety (genetic factors) and climate, especially light. The latter appears to be of greatest influence the last two weeks of ripening time. Excessive heat may possibly have an adverse effect on the ascorbic acid content according to Reid (20).

A number of papers on the effect of processing on the vitamin C content of tomato juice appeared from 1930 to 1935, employing the bioassay method. These reports, notably Kohman, Eddy and Gurin (13) showed experimentally that if tomato juice were aerated during extraction much of the vitamin C was lost. However, if the tomatoes were crushed and boiled before extraction, or if the juice were immediately subjected to a vacuum, losses were small. Daggs and Eaton (7) examined the manufacture of one brand of commercially canned tomato juice reporting that tomato juice may be canned with little or no loss. By the methods employed at that time, it was difficult to detect small losses even if present.

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Recent papers by Tressler and Curran (22), Maclinn and Fellers (16), Fellers and Buck (8), and Hauck (10), employing chemical methods for estimating ascorbic acid, have dealt with certain factors affecting retention of ascorbic acid in preparation and storage of home-processed and experimentally packed tomato juice. Lueck and Pilcher (15) presented information on various factors affecting retention of vitamins and flavour in canned fruit juices. A search of the literature has failed to reveal any comprehensive studies on the effect of processing methods under factory conditions on the retention of ascorbic acid in commercially canned tomato juice or tomatoes.*

A survey in 1942 by the Council on Foods of the American Medical Association (1) of a number of brands of tomato juice canned in the United States, showed a range of from 10 to 28 mg. per 100 grams of juice. The Eastern District (Philadelphia to Buffalo) was low with 10.4 to 13.3 mg., the Central District (Cincinnati to New Orleans) showed 14.9 to 20.7 mg. and the Western District (San Francisco-Denver-Seattle) gave values of 20.7 to 28.0 mg. In 1943 the East and Central Districts showed approximately the same ascorbic acid values and the West was again markedly higher. Another report by the Council on Foods (2) stated that the loss in canning is appreciable in tomato juice and intimated that improvement in the processing of this juice could be anticipated.

MATERIALS AND METHODS

In 1941 studies were commenced by this laboratory on the influence of processing factors on the retention of ascorbic acid in canned tomato juice. Advantage has also been taken of tomato varietal studies to determine the influence of variety on the ascorbic acid content of the canned product. In view of the definite need for maximum retention of vitamin C in commercial canned tomato juice, studies of the various steps in processing

FIGURE 1. Tomato variety test plots at the Dominion Experimental Station, Summerland, B.C.

* See footnote on page 94.

were made under actual operating conditions at several factories in 1943 and 1944. Furthermore, an extensive survey of the ascorbic acid content of canned tomato juice and tomatoes from the three commercial producing areas in Canada, namely, Quebec, Ontario and British Columbia, was made in 1944. This paper presents results of these studies to date.

The tomatoes employed in the study of varietal differences were grown side by side on experimental plots at the Summerland Experimental Station under good cultural and fertilizer conditions on light loam soil. In a number of cases, varieties were grown in the same area over a period of 3 to 4 years. They were harvested at correct canning maturity, prepared and canned in 28-ounce cans in the normal manner, care being taken to exhaust to 160° F. at the centre of the packed can prior to sealing. Two to 3 cans were used for analysis in each case unless otherwise noted. Employing the canned product for determination of the ascorbic acid content of the variety has several advantages, notably from the point of view of convenience and reduction of sampling error. The loss of ascorbic acid on canning under good conditions is insignificant and is comparable for all varieties being tested. Replicate cans were also examined to determine the canning characteristics of each variety. Results of these tests will be reported later.

In testing the influence of processing methods on the ascorbic acid content of tomato juice, 7 or more pounds of tomatoes, usually Earliana 8040, were used to prepare each lot of juice. The basic method of preparation was that described by Atkinson and Strachan (3) for home canned tomato juice. A stainless steel steam jacketed kettle was used for heating the tomatoes and juice to desired temperatures.

In 1944, in co-operation with the Canning Division of the Marketing Service, Dominion Department of Agriculture, 318 samples of tomato juice and 88 samples of canned tomatoes were obtained from the three commercial producing areas in Canada (Quebec, Ontario and British Columbia). These samples were analysed for ascorbic acid and proximate chemical composition and were examined for flavour, colour and other characteristics. Notes were also made of net weight of contents, head space, and vacuum. Only ascorbic acid determinations are being reported in this paper.

Ascorbic acid (reduced) was determined by the sodium 2,6-dichlorophenol indophenol dye visual titration method of Bessey and King (4), employing a 5- to 7-second end point. Daylite fluorescent light and white base were used to increase the accuracy of the end point determination. The dye was standardized according to the method of Buck and Ritchie (5). Occasionally standardization was carried out against pure crystalline ascorbic acid, good agreement being obtained by both methods. The extractions were made with 2% metaphosphoric acid or 0.4% oxalic acid (Ponting, 18).

For juice, 50 ml. were pipetted into a 250-ml. volumetric flask containing extractant, made up to volume with acid extractant, shaken thoroughly and filtered. Five to 15 ml. aliquots were then titrated rapidly with the dye and results reported as mg. per 100 ml. of juice. Where necessary, for certain comparative purposes, the determinations were recalculated to weight basis from the specific gravity of the juice. In nearly all tests, samples were taken in duplicate or triplicate.

For canned tomatoes, determinations were carried out as for juice on 50-gram portions after mixing of the whole contents of the can in a Waring blendor for 30 seconds. This operation permitted more accurate sampling. Tests showed no ascorbic acid was lost by this procedure. In fact, blending 3 to 5 minutes resulted in no decrease of ascorbic acid. Unless otherwise noted 2 or 3 canned samples were examined from each lot or test.

Fresh fruit was prepared for analysis by cutting quarters from 4 fruits to make 100 grams and extracted with 400 ml. of oxalic or metaphosphoric acid extractant in a Waring blendor for 2 minutes and filtered through No. 4 or No. 12 Whatman filter paper. Five to 15 ml. aliquots of the filtrate were titrated with the dye. This method is essentially that described by Loeffler and Ponting (14). The procedure was repeated three or more times for any sample of fruit so that at least 12 representative fruits and usually more were used for each sample, the mean of the individual determinations being recorded.

Tests for interference of metallic ions in the canned product to the dye titration showed that there was none.

RESULTS

The Effect of Variety and Season

In Tables 1 and 2 are presented the ascorbic acid values for 31 different varieties and strains of tomatoes grown under the same conditions at the Summerland Experimental Station. It will be noted from these tables that there was marked variation in ascorbic acid content of varieties. Also while there was considerable variation in ascorbic acid values in the same variety from year to year and even from one picking to another, the varieties tended to maintain their relative position of one to another especially where average difference was marked. These results are in general agreement with the current literature on this question. The Signet variety developed at the Summerland Station is outstanding in its consistently high ascorbic acid content having a 3-year average of 29.8 mg. of ascorbic acid. Its only serious fault is that it lacks size for a good commercial canning tomato. Clarks Early and Sugawara have proved to have consistently good ascorbic acid values but are not outstanding. Fortunately, Clarks Early and Sugawara have good cultural and canning characteristics which make them satisfactory canning varieties under actual commercial conditions in the Okanagan Valley and adjacent areas. Harkness and Bestal (Sd.) have shown consistently fairly good ascorbic acid values but are not satisfactory for other reasons. Master Marglobe, Marglobe × Bonny Best, Stokesdale, Valiant, Livingstone Globe × Gnome and Essary, while having on limited trial apparently good ascorbic acid content, are unsatisfactory in other respects, either not suited to this growing area or else unsatisfactory for canning. California Dawn, a good sized tomato and a satisfactory canner, had on 2 years tests a fairly good ascorbic acid content.

In general, the ascorbic acid contents of the varieties recorded in Tables 1 and 2 compare favourably with the higher results for the same varieties reported in the literature. These high values are probably due in part to favourable climate with respect to temperature and light as

indicated in studies by Reid (19, 20, 21), Wokes and Organ (23) and Kaski, Webster and Kirch (11). It is interesting to note from Table 1 that under conditions prevailing, there was no consistent difference in the ascorbic acid content of tomatoes harvested early and late in the season.

TABLE 1.—ASCORBIC ACID CONTENT OF VARIETIES AND STRAINS OF TOMATOES HARVESTED AT SEVERAL DATES OVER A PERIOD OF THREE TO FOUR YEARS

Variety or strain	Ascorbic acid content in milligrams per 100 grams							Average for 3 or 4 years
	Dates tomatoes harvested and canned							
	1941	1942	1943			1944		
	Sept. 4	Sept. 22	Sept. 10	Sept. 29	Oct. 19	Sept. 6	Sept. 27	
	mg.	mg.	mg.	mg.	mg.	mg.	mg.	mg.
Signet	33.1	—	23.7	29.1	34.9*	24.4	33.7	29.8
Clarks Early	—	31.7	20.9	24.1	23.2*	—	18.9	23.8
Harkness Early	24.7	19.7	24.2	25.1	—	—	—	23.4
Sugawara	24.5	26.3	22.8	25.7	22.0*	20.4	19.0*	23.0
Bestal (Sd.)	23.3	24.7	20.8	21.2	—	—	—	22.5
Dick Locke (Round)	—	22.1	17.4	23.0	23.4	20.5	20.0†	21.1
Signet × Clarks Early (Sd.)	16.4	24.6	20.1	21.5	—	—	—	20.7
Sentinel (Sd.)	—	24.1	16.0	21.7	16.6*	21.4	22.8	20.4
Bounty	16.6	20.7	—	—	—	15.8*	20.8	18.5

* One sample only.
† Another sample from quite different soil (Trout Creek) contained 22.8 mg. ascorbic acid.

TABLE 2.—ASCORBIC ACID CONTENT OF TWENTY-TWO VARIETIES AND STRAINS OF TOMATOES

Variety or strain	Ascorbic acid content in milligrams per 100 grams of canned product			
	1941	1942	1943	1944
	mg.	mg.	mg.	mg.
Master Marglobe (Stokes)	30.5	—	—	—
Marglobe × Bonny Best	30.2	—	—	—
Signet × Sugawara	—	—	—	30.0
Stokesdale (Stokes)	29.3	—	—	—
Valiant (Stokes)	28.3	—	—	—
Sugawara × Signet	—	—	28.1	27.5*
Asgrow Scarlet Dawn	—	—	—	23.8*
Globonnie	26.3	—	—	—
Livingstone Globe × Gnome × L. G. × Abel	26.2	—	—	—
Essary (Univ. of Tenn.)	25.5	—	—	—
Hybrid 46 (Mich.)	24.4	—	—	—
California Dawn	—	23.6	22.6†	—
8040 (Earliana Sport)	—	25.1	21.2*	—
Signet × John Baer	—	—	—	21.8*
Rutgers (Vineland)	21.5	—	—	—
Rutgers (Stokes)	21.5	—	—	—
Signet × California Dawn	—	—	—	21.7*
Abel	19.9	—	—	—
Bulman's Special (Flat)	—	18.8	20.9†	—
N.D.A.C.	17.9	—	—	—
Bison	17.3	—	—	—
Bulman's Flat (Sd. 993)	—	—	23.0*	15.2

* Average of two pickings.
† Average of three pickings.

The production of a high vitamin C, good quality, heavy yielding tomato with satisfactory canning and juice characteristics appears to be a very urgent problem for the plant breeder. In this connection it is interesting to note that crosses made at the Summerland Station, using the high vitamin C variety Signet as one parent, have given encouraging results. Data presented in Table 3 indicate the possibilities of developing a variety combining high ascorbic acid content with superior cultural and canning characteristics. These data were obtained by analysing fruit from individual plants selected from among many in breeding plots at the Experimental Station. These plants were originally selected on the basis of plant and fruit characteristics suitable for commercial market or cannery production.

TABLE 3.—ASCORBIC ACID CONTENT OF TOMATOES FROM INDIVIDUAL PLANTS, 1943

Plant No.	Parentage	Ascorbic acid content mg. per 100 gm.
		mg.
1–43	John Baer × Signet	34.2
2–43	John Baer × Signet	23.9
3–43	Signet × Calif. Dawn	31.0
4–43	Signet × Calif. Dawn	20.8
5–43	Signet × Calif. Dawn	34.4
6–43	Sugawara × Signet	35.9
7–43	Sugawara × Signet	41.6
8–43	Signet × Sugawara	32.4
9–43	Signet × Sugawara	34.8

Ascorbic Acid Content of Commercially Canned Juice and Tomatoes

The results of a survey of the ascorbic acid values of tomato juice and canned tomatoes as commercially produced are presented in Table 4. There is considerable variation in the ascorbic acid content of tomato juice with somewhat less variation in values for canned tomatoes. However, the most important points to note in this table are: (1) the significantly higher average ascorbic acid content of 22.3 and 19.8 mg. for tomato juice manufactured in British Columbia compared with 14.4 and 15.0 mg. for Eastern Canada; (2) the relatively high minimum values of 12.6 and 15.6 mg. for British Columbia produced tomato juice and the low minimum values of 8.7 and 4.3 mg. for Eastern packed juice while the maxima of both districts are more nearly comparable; (3) the minimum values for ascorbic acid in canned tomatoes are almost identical for both Eastern Canada and British Columbia. These results suggest that many of the low values for tomato juice are to a large extent due to faulty processing and unsatisfactory equipment. The average ascorbic acid values in canned tomatoes from widely separated districts suggest inherent differences which are probably due largely to climate.

TABLE 4.—ASCORBIC ACID CONTENT OF COMMERCIALLY CANNED TOMATOES AND TOMATO JUICE IN CANADA

Year packed	Area	No. of samples analysed	Ascorbic acid values per 100 ml. of juice		
			Average	Maximum	Minimum
			mg.	mg.	mg.
JUICE					
1941	British Columbia	18	22.3	33.3	12.6
1942	Eastern Canada	12	14.4	19.5	8.7
1944	Quebec	44	14.2	26.0	6.1
	Ontario	220	15.1	25.0	4.3
	British Columbia	54	19.8	26.9	15.6
	Eastern Canada	264	15.0	26.0	4.3
TOMATOES			per 100 grams of tomatoes		
1940–41	British Columbia	39	21.6	32.4	14.4
1944	Quebec	5	15.9	18.9	14.1
	Ontario	30	17.2	20.3	15.1
	British Columbia	53	22.5	27.7	16.3
	Eastern Canada	35	17.0	20.3	14.1

Comparison of Ascorbic Acid Content of Juice and Tomatoes Canned Commercially

In order to ascertain how the retention of ascorbic acid in tomato juice compared with that of tomatoes, the data in Table 5 were compiled. The factory from which the samples were secured was well equipped and considered to be packing both products satisfactorily. Canned samples of juice and tomatoes were taken on the same day and also at the same hour when possible. The canned tomato samples obviously represent smaller numbers of fruit than do the juice samples but it is believed there are sufficient samples to make the results significant. Under good processing conditions it would appear that on the average the tomato juice contains about 14% less ascorbic acid than do comparable canned tomatoes. A limited survey of samples from a number of other factories indicates that losses much greater than this occur. Indications are that tomatoes lose very little ascorbic acid in canning. Unpublished data of the Chemistry Division, Science Service, Ottawa (6), showed no significant loss of ascorbic acid in commercial canning of tomatoes.

Studies on Retention of Ascorbic Acid in Tomato Juice Under Factory Conditions

In 1943 it was decided to study ascorbic acid retention under commercial conditions by following the produce as received at the factory through the various processing steps in the plant. This survey was carried out in one plant that year with certain recommendations being made as to improvements in the manufacturing line. The study was repeated in 1944 at the same factory as well as at two additional factories. The results

TABLE 5.—ASCORBIC ACID CONTENT OF CANNED TOMATOES AND TOMATO JUICE PACKED ON SAME DAY AT FACTORY C

(1944 season)

Sample number	Date packed	Ascorbic acid content per 100 grams	
		Juice	Tomatoes
		mg.	mg.
1	Sept. 14	16.7	18.5
2	Sept. 15	21.3	24.3
3	Sept. 19	20.3	22.9
4	Sept. 21	15.2	27.7
5	Sept. 22	21.5	18.9
6	Sept. 23	17.2	21.1
7	Sept. 25	22.5	23.2
8	Sept. 26	22.5	17.8
9	Sept. 27	18.6	21.7
10	Sept. 28	20.2	24.3
11	Sept. 30	19.1	21.1
12	Sept. 30	19.3	—
13	Oct. 2	21.4	23.2
14	Oct. 3	23.5	27.1
15	Oct. 5	23.2	23.4
16	Oct. 6	17.2	23.2
17	Oct. 7	21.2	22.2
18	Oct. 11	20.1	22.2
19	Oct. 13	18.4	20.9
20	Oct. 16	18.1	19.8
21	Oct. 16	15.7	—
22	Oct. 17	26.1	31.6
23	Oct. 18	17.0	24.4
Average		19.8	22.8
Maximum		26.1	27.7
Minimum		15.2	17.8

of the study at Factory C in 1944 are given in Table 6. This factory has what is considered to be a very good tomato juice manufacturing line. Losses were in general smaller and less variable at this plant than in the other two plants investigated. The ascorbic acid figures recorded in Table 6 for raw fruit are very approximate due to the fact that only 12 to 16 fruits were analysed whereas the other figures represent a volume of 50 to 100 gallons of juice or equivalent to around 700 to 1400 pounds of tomatoes. It is unlikely that there was a significant loss at this plant in the hot break due to the few seconds only required following milling to reach the inactivating temperature of 190° F. Hence this figure probably should be taken as representing more nearly the true average ascorbic acid value of the raw product being employed during the tests. On this basis the total loss in processing under good conditions was 2 to 3 mg. or 11.7 to 13.5%. Loss at Factory A was only very slightly greater than at C, but Factory B showed a loss of 18.1 to 26.9%. This was probably due to low temperature extraction allowing enzyme action together with aeration of the product. Losses greater than those found could be expected under such conditions. None of the factories studied permitted contact of the product with copper equipment.

TABLE 6.—RETENTION OF ASCORBIC ACID AT PROGRESSIVE STEPS IN COMMERCIAL PROCESSING OF TOMATO JUICE

Test No.	Steps in processing	Ascorbic acid per 100 gm.
		mg.
	FACTORY C (1944)	
A 1	Raw tomatoes	25.0
2	Emerging from hot break at 200° F.	22.2
3	In finisher receiving tank at 188-190° F.	20.7
4	Holding-salting tank (100 gal.) at 183° F.	19.4
5	After filler prior to sealing can at 182° F.	19.3
6	After canned juice stored three weeks	19.2
B 1	Raw tomatoes	20.8
2	Emerging from hot break at 200° F.	19.6
3	In finisher receiving tank at 188-190° F.	19.2
4	Holding-salting tank (100 gal.) at 183° F.	17.2
5	After filler prior to sealing can at 182° F.	16.3*
6	After canned juice stored three weeks	17.3

* Froth at time of sampling, indicating air in product, likely accounts for this low figure. More frothing seemed to occur with the less mature fruit.

Effect of the Method of Extraction on the Ascorbic Acid Content

Laboratory experiments were carried out to determine the effect of different temperatures employed in preheating the tomatoes prior to extraction and also the actual method of extraction on the ascorbic acid content of the resultant canned juice. The results obtained are reported in Table 7. All tests were made from the same lot of raw tomatoes; 7 to 10 pounds of tomatoes were used for each test. In test No. A, B, J, and K, the tomatoes were placed in a small stainless steel jacketed kettle, pulped while heating in about 5 minutes to 210° F. (boiling at this altitude) using 20-25 lb. steam pressure, boiled specified time, then extracted by various methods. To pass the cooked tomato pulp through the suspended screen by hand required about 3 minutes. Tests C and D were pulped and heated as for A but to lower temperatures. In test E, whole tomatoes were placed in boiling water, and in test F, whole tomatoes were exposed to flowing steam. In tests G, H, and I, whole fruit was extracted without any heating. In all cases, the juice obtained by the various methods of extraction was heated quickly in the kettle to 190° F. and the cans filled full at that temperature, sealed, processed in boiling water 10 minutes and then water cooled.

The very important point brought out in Table 7 is the necessity of rapid heating of the milled pulp to sufficiently high temperature (at least 190° F.) to inactivate quickly the relatively strong oxidase enzymes present in the tomatoes. If this is done, loss of ascorbic acid is relatively small but if not, the loss may be serious as indicated in Table 7 where as much as 36% loss occurred. The actual method of extraction appeared to be of little consequence provided the extraction was on pulp heated to 190° to 210° F. This temperature has the additional advantage that it also inactivates pectin-destroying enzymes resulting in improved consistency of juice as pointed out by Kertesz and Loconti (12).

TABLE 7.—EFFECT OF THE METHOD OF EXTRACTION ON THE ASCORBIC ACID CONTENT OF EXPERIMENTALLY PREPARED CANNED JUICE

Test No.	Method of extraction	Ascorbic acid per 100 ml.	Per cent loss compared to Lot K
		mg.	%
A	Pulped and boiled 3 to 4 min. Extracted hot through screen	30.2	8.5
B	Pulped and boiled ½ to 1 min. Extracted hot through screen	29.6	10.3
C	Pulped and heated to 173° F. Extracted hot through screen	22.4	32.1
D	Pulped and heated to 110° F. Extracted immediately	22.9	30.6
E	Scalded whole 3 min. in boiling water, pulped and extracted immediately through screen	24.5	25.8
F	Scalded whole 2 min. in flowing steam, pulped and extracted immediately through screen	22.9	30.6
G	Pulped and cold extracted through screen	22.5	31.8
H	Pulped and cold extracted through centrifugal juicer	21.8	33.9
I	Pulped and cold extracted through screw expeller press	21.0	36.4
J	Pulped and boiled ½ to 1 min. Extracted hot through small centrifugal juicer	25.2	23.6
K	Pulped and boiled ½ to 1 min. Extracted hot through small screw expeller press	33.0	0.0

Effect of Sterilizing Temperature and Period

To study the effect of sterilizing temperature and time on the ultimate ascorbic acid content of canned tomato juice, several lots of juice were prepared according to Atkinson and Strachan (3) home process procedure. The results of this experiment are recorded in Table 8. The cans in each lot were filled full at 190° F., sealed, sterilized as recorded in the table, and water cooled. Analyses were made after several months storage at 50° F. One-half the cans from each lot of prepared juice were held hot on their sides in the air for 5 minutes or processed in boiling water for 10 minutes for controls. The different lots were not always necessarily prepared from the same fruit. The results show that the sterilizing temperature had very little effect on the final ascorbic acid content of canned tomato juice. This is in agreement with limited data obtained in 1943 under commercial conditions. It was observed that excessive processing temperatures and long processing time had a deleterious effect upon colour and flavour.

SUMMARY

Analyses for ascorbic acid content have been made of 31 varieties and strains of tomatoes grown under identical conditions at the Summerland Experimental Station. A number of the varieties were examined over a period of 3 to 4 years. Clarks Early and Sugawara were found to have consistently good ascorbic acid values and are satisfactory canning tomatoes in this area. The Signet variety proved to be consistently high in ascorbic acid, having a mean value of 29.8 mg. per 100 grams over a 3-year period. Its other characteristics are also good with the exception that it tends to be small. The fruits from individual plants, resulting from crossing Signet with larger fruited varieties, were analysed for ascorbic acid. The results indicate the practicability of developing varieties combining high ascorbic acid content with superior cultural and canning characteristics.

TABLE 8.—EFFECT OF STERILIZING TEMPERATURE AND PERIOD ON ASCORBIC ACID CONTENT OF EXPERIMENTALLY CANNED TOMATO JUICE

Lot No.	Processing data		Ascorbic acid per 100 ml.
	Sterilizing process	Period	
		min.	mg.
1 (a)	210° F. (Boiling water)	10	26.2
(b)	(Held on side in air)	5	26.5
2 (a)	240° F. (Retort)	10	26.6
(b)	210° F. (Boiling water)	10	26.3
3 (a)	240° F. (Retort)	20	27.9
(b)	210° F. (Boiling water)	10	29.3
4 (a)	250° F. (Retort)	5	24.9
(b)	210° F. (Boiling water)	10	25.9
5 (a)	250° F. (Retort)	15	26.8
(b)	210° F. (Boiling water)	10	28.0

A comprehensive survey was made of commercially canned juice and tomatoes in Canada. Marked differences were found in average ascorbic acid content of tomato juice produced in British Columbia and Eastern Canada. In 1941 and 1944 the British Columbia mean values were 22.3 and 19.8 mg., respectively, while the 1942 and 1944 values for Eastern Canada were 14.4 and 15.0 mg. Very low values were found in tomato juice packed in Eastern Canada, yet these low values were not found in the canned tomato samples.

An extensive study was made at three factories of the retention of ascorbic acid in tomato juice under factory conditions. Analyses were made at several steps in the process of manufacture. This study revealed that under good processing conditions the total loss in processing from the raw fruit to the final canned product should not exceed 2 to 3 mg. Loss at many factories apparently greatly exceeds this figure.

Laboratory experiments were conducted on the effect of the method of extraction and the effect of sterilizing temperatures and time on the retention of ascorbic acid in canned tomato juice. The great importance of rapidly heating the milled tomatoes to 190° F.–210° F. prior to extraction was demonstrated. The sterilizing temperature and length of cook had insignificant effect on the ultimate ascorbic acid content of the canned juice. Excessive sterilizing did, however, adversely affect the colour and flavour of the juice.

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* Since the preparation of this paper, two studies have been reported on factors influencing ascorbic acid retention in commercially processed tomato juice: (1) Clifcorn, L. E., "Variables influencing vitamin content of processed foods," The Food Packer, pp. 46-48, August, 1945, and (2) A Memorandum by the Research Department of the American Can Company on "Tomato juice—factors influencing ascorbic acid retention," 1945.

PRECANNING TREATMENT ON PROCESSED PLUMS AS IT AFFECTS QUALITY AND VITAMIN C CONTENT[1]

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Preprocessing by scalding in water containing bicarbonate of soda has been generally recommended to remove the acridness present in the skin of most native plums. This practice has been condemned by nutritionists and home economists. The reason for criticism was the belief that the Vitamin C content of the processed fruit was reduced.

During experimental work on fruit varietal quality, at this Station, a study of the effect of preprocessing with bicarbonate of soda, sodium chloride, lye and boiling water on the Vitamin C content of plums was undertaken. Additional treatments were made in an endeavour to find a process that would not reduce the nutritive value and yet would give an attractive, palatable product. Fourteen varieties were used in the test.

Pett and Cantor (1) state that although Vitamin C is the least stable of the vitamins it is quite stable when canned in some foods because of the presence of combined forms. They showed that the Vitamin C content of prunes, dried or canned, is very low. Investigations by Tuba *et al.* (4) on the loss of ascorbic acid in processed native fruits over a period of from 5 to 7 months indicated variations of 0% to 75%. Tuba *et al.* (2), reported in their ascorbic acid determinations of rose hips a technique for preparing materials, using metaphosphoric acid as a fixative and 2,6-dichlorophenol-indophenal for titration. The Vitamin C content of *Prunus melanocarpa* and *Prunus pennsylvanica* as reported by Tuba *et al.* (3) varied from 5 mg. % to 30 mg. %. Hunter and Tuba (unpublished data) found the ascorbic acid value of the Assiniboine plum, a variety comparable with those reported on here, to be 2 mg. %, which is very low when considered from a nutritional standpoint.

EXPERIMENTAL

Varieties of mature plums were washed, pricked, and covered with a thin syrup (1 cup sugar, 2 cups boiling water) and processed in a boiling water bath in number one enamel lined cans for 10 minutes. Processing time was taken from the time the water boiled after the cans were immersed.

Duplicate cans of standard and of each preprocessing treatment were prepared. These treatments were: (a) the fruit was washed, pricked, covered with boiled syrup and sealed; (b) the fruit was washed, pricked and allowed to stand for 5 minutes in boiling water, covered with syrup, and sealed; (c) the fruit was washed, pricked, and allowed to stand for 5 minutes in a hot bicarbonate of soda solution (1.34 gm. ($\frac{1}{2}$ teaspoon) soda in one pint of boiling water) and was then rinsed, covered with boiled syrup and sealed; (d) the fruit was washed, pricked, and allowed to stand for 3 minutes in a hot lye solution (11 mg. of lye (commercial NaOH) in

[1] Contribution from the Provincial Horticultural Station, Brooks, Alberta.
[2] Superintendent.
[3] District Home Economist, Calgary, Alberta.

in one pint of boiling water) which was removed by rinsing, and the fruit was then covered with boiled syrup and sealed; (e) the fruit was washed, pricked, and allowed to stand for 3 minutes in hot saline solution (5.56 gm. (1 teaspoon) of sodium chloride in one pint of boiling water), rinsed, and then covered with boiled syrup and sealed.

Fruit canning quality results were based on score. The score guide evaluated the fruit as to form, colour, skin texture, flesh texture, skin flavour, flesh flavour, juice flavour, and sweetness.

Fruit Quality Score Guide

Form 1-10	10 indicates perfect.
Colour 1-10	10 indicates bright, clean, attractive.
Skin texture 1-10	1 is tough; 10 is tender and thin; above 5 is tender enough to be palatable.
Flexh texture 1-10	1 is soft, mushy, soapy or unpleasantly coarse and stringy; 10 is firm, smooth pleasing.
Skin flavour 1-20	1 is bitter and sour; 20, no bitterness and pleasant.
Flesh flavour 1-20	1 is sour, bitter; 20 aromatic, pleasant, tasty.
Juice flavour 1-10	1 is soft, mushy, soapy or unpleasantly coarse and stringy; 10 is firm, smooth, pleasing.
Sweetness 1-10	1-5 is too sour or too sweet with prefix A for sour, B for sweet; 6-10 desirably sweet as 10 is approached.

The cans were stored at 40° F. for 4 months. Duplicate cans were opened and scored by a testing team of 2 persons. Immediately upon opening the cans, the portions used for Vitamin C determinations were treated with metaphosphoric acid to prevent deterioration of the ascorbic acid.

Determination of Vitamin C Content

The cans were opened and the fruit immediately prepared for ascorbic acid determination; 9 ml. of plum pulp containing skin and 6 ml. of juice were mixed in a Waring Mixer for 2 minutes. Pits were eliminated before mixing the pulp and juice.

Thirty ml. of aqueous extract consisting of 15 ml. of H_2O plus 15 ml. of blended fruit product were used for making the determinations. Ten ml. of 2% metaphosphoric acid ground in 2N hydrochloric acid was added and blended giving 40 ml. of mixture. This was transferred to a graduate centrifuge tube and centrifuged. Precise aliquots of the supernatant fluid were titrated with 2,6-dichlorophenolindophenol.

RESULTS

The varieties tested are listed in Table 1, which shows the standard, standard and boiling water, standard and soda, standard and lye, and standard and salt treatment with the quality scored in percentage under each method and the vitamin C content expressed in mg. %. The methods

are listed in their descending order of average vitamin C content. This does not hold completely throughout the table as there are instances where the soda and lye preprocessing treatment showed a slight improvement in score over the standard method. The two hybrids Opata and Sapa were most markedly affected in this regard. Treatment with bicarbonate of soda and lye accentuated the sweetness in Opata and resulted in a higher total score. Sweetness score for Sapa was constant with the same treatments. The results from a canning standpoint showed that the standard method gave the highest average score for quality and, as was to be expected, retained the greatest vitamin C content. All methods of preprocessing reduced the ascorbic acid content and the quality with a few exceptions was similarly impaired. Varieties with skins typical of *Prunus nigra* were improved in quality or held their own by preprocessing with boiling water.

TABLE 1.—FRUIT VARIETY SCORE AND VITAMIN C CONTENT*

Variety	Standard		Standard and B. water		Standard and soda		Standard and lye		Standard and salt	
	Score	Vitamin content	Score	Vitamin content	Score	Vitamin content	Score	Vitamin content	Score	Vitamin content
Bounty	66	.73	66	.47	68	.33	64	.27	60	.40
Brooks 41	68	.60	65	.52	64	.40	63	.40	—	—
Ember	70	2.60	68	1.80	68	2.20	68	1.27	66	1.73
Etopa	71	1.40	70	.93	77	1.73	74	1.60	72	1.47
Emerald	77	2.13	77	.93	77	1.73	74	1.60	72	1.47
La Crescent	88	.67	82	.60	85	.47	83	.40	80	.47
Louise	75	1.50	74	1.00	74	1.13	65	.75	—	—
McRobert	70	.87	69	.73	69	.60	68	.67	65	.67
Mina (M 109)	61	1.13	66	.73	61	.40	64	.40	—	—
Minn. 255	84	2.00	82	.81	82	.69	82	.63	—	—
Opata	84	.73	84	.67	86	.33	86	.40	—	—
Red Wing	77	1.07	70	.52	72	.33	72	.40	69	.60
Sansota	71	.73	64	.52	69	.47	64	.33	—	—
Sapa	91	2.06	86	1.40	91	1.27	91	.80	—	—
Average	75.2	1.3	73.0	0.8	74	0.8	72.5	0.6	68.2	0.8

* 100% perfect score. Vitamin content expressed in mg. %.

DISCUSSION

The evidence presented indicates that preprocessing treatments of boiling water, soda, lye and salt impair the quality of the final product. This contradicts the widely held belief that such treatments are beneficial.

All treatments used reduce the vitamin C content to a greater extent than does the standard method. The reduction of the ascorbic acid content is more marked with bicarbonate of soda and lye than are pretreatments with boiling water or salt.

Preprocessing treatments are not recommended, but if one is to be used a mild sodium bicarbonate solution is preferred because of the tendency to give a better final product. The vitamin C content of this final product might be slightly lower than when a boiling water pretreatment is used.

SUMMARY

Preprocessing of native plums with boiling water, soda, lye and salt reduces ascorbic acid content. The treatments do not improve the palatability of the processed product and recommendations of this effect are not advisable.

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